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MAY 10 AND 11, 1921

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The Association assumes no responsibility for the statements and opinions expressed in the papers read at its meetings.

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1897.	1905.	Cushny, Arthur C., University of Edinburgh, Edinburgh, Scotland.
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*1886.	1905.	Edes, Robert T., 340 Summer Ave., Reading, Mass.
1901.	1919.	Edwards, A. R., 104 S. Michigan Ave., Chicago.
1888.	1919.	Ernst, H. C., 8 Greenough Ave., Jamaica Plain, Mass.
1890.	1902.	Fisk, S. A. Brimfield, Mass.
*1886.	1900.	Guitéras, John, Havana, Cuba.
*1886.	1902.	Hays, I. Minis, 268 S. 21st St., Philadelphia.
1897.	1910.	Hektoen, Ludvig, 637 S. Wood St., Chicago.
1900.	1917.	Holt, L. E., 14 W. 55th St., New York.
1890.	1902.	Hurd, H. M. Johns Hopkins Hospital, Baltimore.
1899.	1914.	James, Walter B., 17 W. 54th St., New York.
1895.	1918.	Lafleur, H. A., 215 Peel St., Montreal, Canada.
1888.	1919.	Lewis, M. J., 1316 Locust St., Philadelphia.
1891.	1920.	McPhedran, A., 151 Bloor St., W., Toronto, Can.
1898.	1916.	Mallory, F. B., Harvard Medical School, Boston.
1891.	1900.	Mason, A. Lawrence, 265 Clarendon St., Boston.
1901.	1913.	Morse, John L., 70 Bay State Road, Boston.
1891.	1918.	Northrup, W. P., 61 E. 73d St., New York.
1904.	1920.	Pearce, R. M., Rockefeller Foundation, 61 Broadway, New York.

ELECTED ACTIVE MEMBER	ELECTED EMERITUS MEMBER	
1898.	1914.	Pfaff, Franz, Dresdner Bank, Berlin.
*1886.	1898.	Polk, Wm. M., 7 East 36th St., New York.
1903.	1918.	Preble, R. B., 590 Dearborn Ave., Chicago.
*1886.	1904.	Robinson, Beverley, 42 West 37th St., New York.
1900.	1914.	Sears, Geo. G., 19 Marlborough St., Boston, Mass
1900.	1921.	Sewall, Henry, 1360 Vine St., Denver, Col.
*1886.	1912.	Shattuck, Frederick C., 135 Marlborough Street, Boston.
*1886.	1905.	Shattuck, George B., 183 Beacon St., Boston.
1887.	1911.	Starr, M. Allen, 5 West 54th St., New York.
1907.	1920.	Taylor, A. E., Univ. of Penna., Philadelphia.
1899.	1918.	Thacher, J. S., 20 W. 50th St., New York.
1898.	1918.	Vickery, H. F., Ipswich, Mass.
1889.	1904.	Walcott, Henry P., 11 Waterhouse St., Cambridge, Mass.
1894.	1920.	Williams, F. H., 505 Beacon St., Boston.
*1886.	1911.	Wilson, James C., 1509 Walnut St., Philadelphia.
1899.	1912.	Wright, J. H., Pathological Laboratory, Massachusetts General Hospital, Boston.

DECEASED MEMBERS

* Original Members

DATE OF ELECTION

1896. Adler, Isaac, New York.
*1886. Atkinson, I. E., Baltimore.
*1886. Ball, A. Brayton, New York.
1890. Barker, Fordyce, New York.
*1886. Baumgarten, G., St. Louis.
*1886. Billings, John S., New York.
*1886. Bowditch, Henry I., Boston.
*1886. Bruen, Edward T., Philadelphia.
*1886. Busey, Samuel C., Washington, D. C.
1907. Carroll, James, Washington, D. C.
*1886. Chew, Samuel C., Baltimore.
*1886. Clark, Alonzo, New York.
*1886. Clymer, Meredith, New York.
*1886. Da Costa, J. M., Philadelphia.
1889. Dabney, William C., University of Virginia.
*1886. Dalton, John C., New York.
*1886. Dana, Israel T., Portland, Maine.
1888. Danforth, I. N., Chicago.
*1886. Delafield, Francis A., New York.
*1886. Donaldson, Frank, Baltimore.
*1886. Draper, William H., New York.
1899. Ely, John Slade, New Haven, Conn.
1895. Fischel, William E., St. Louis.
*1886. Fitz, Reginald H., Boston.
*1886. Folsom, Chas. F., Boston.
*1886. Forchheimer, F., Cincinnati.
*1886. Formad, Henry F., Philadelphia.
1889. Geddings, W. H., Atken, S. C.
1916. Gorgas, W. C., Washington.
*1886. Graham, J. E., Toronto, Canada.
*1886. Henry, F. P., Philadelphia.
1898. Herter, C. A., New York.
*1886. Howard, R. Palmer, Montreal, Canada.

DATE OF ELECTION

*1886. Hudson, Jr., E. Darwin, New York.
*1886. Hutchinson, James H., Philadelphia.
*1886. Jacobi, A., New York.
*1886. Janeway, E. G., New York.
1908. Janeway, T. C., Baltimore.
*1886. Johnson, Hosmer A., Chicago.
1899. Johnston, W. G., Montreal, Canada.
*1886. Johnston, W. W., Washington, D. C.
1902. Kelly, A. O. J., Philadelphia.
*1886. Kinnicutt, Francis P., New York.
1901. Kinyoun, J. J., Washington.
1890. Latimer, Thomas S., Baltimore.
*1886. Leidy, Joseph, Philadelphia.
*1886. Longstreth, Morris, Cambridge, Mass.
*1886. Loomis, Alfred L., New York.
1889. Loving, Starling, Columbus, O.
*1886. Lyman, Henry M., Chicago.
*1886. McBride, Thomas A., New York.
1914. McCrae, John, Montreal, Canada.
1890. MacDonnell, R. P., Montreal, Canada.
1887. Meigs, Arthur V., Philadelphia.
1892. Meltzer, S. J., New York.
*1886. Metcalfe, John T., Cold Springs, N. Y.
*1886. Miles, F. T., Baltimore.
*1886. Minot, Francis, Boston.
1898. Mitchell, John K., Philadelphia.
*1886. Mitchell, S. Weir, Philadelphia.
1887. Musser, John H., Philadelphia.
*1886. Osler, Sir William, Oxford.
1898. Packard, Frederick A., Philadelphia.
*1886. Peabody, George L., Newport, R. I.
*1886. Pepper, William, Philadelphia.
1888. Prentiss, D. W., Washington, D. C.
*1886. Putnam, James J., Boston.
1896. Reed, Walter, Washington, D. C.
1887. Reeves, James E., Chattanooga, Tenn.
*1886. Rochester, Thomas F., Buffalo, N. Y.
1889. Roosevelt, J. West, New York.
*1886. Ross, George, Montreal, Canada.
1893. Rotch, Thomas M., Boston.
*1886. Schmidt, H. D., New Orleans.
*1886. Sinkler, Wharton, Philadelphia.
*1886. Smith, A. A., New York.

DATE OF ELECTION

1891. Smith, Andrew H., New York.
1915. Southard, E. E., Boston.
1905. Steele, J. Dutton, Philadelphia.
*1886. Sternberg, Geo. M., Washington.
1896. Stewart, D. D., Philadelphia.
*1886. Stewart, James, Montreal, Canada.
*1886. Stillé, Alfred, Philadelphia.
*1886. Thacher, James K., New Haven, Conn.
1889. Thomson, W. H., New York.
*1886. Trudeau, Edward L., Saranac Lake.
*1886. Tyson, James, Philadelphia.
*1886. Ward, Samuel B., Albany.
1898. Wesbrook, F. F., Vancouver.
*1886. Whittaker, James T., Cincinnati.
*1886. Wilkins, George, Montreal.
1898. Withington, Charles F., Boston.
*1886. Wood, H. C., Philadelphia.

ASSOCIATION
OF
AMERICAN PHYSICIANS

CONSTITUTION

As Amended May 10, 1921

ARTICLE I

OBJECT AND NAME

This Association has for its object the advancement of scientific and practical medicine, and shall be known as The Association of American Physicians.

ARTICLE II

MEMBERS

The number of active members shall be limited to two hundred. Physicians of sufficient eminence to merit the distinction may, to a number not exceeding twenty-five, be elected honorary members, and as such shall be entitled to attend all meetings and to take part in the proceedings, but not to vote.

An active member whose membership has extended throughout ten years or more may, at his request, be transferred by the Council to emeritus membership.

There shall be also a class of associate members, limited in number to twenty-five, from which class active members may be elected. These associate members shall each be elected for a term of five years and shall enjoy all the privileges of emeritus members, and each of them shall pay annual dues amounting to one-half the annual dues of an active member.

Membership shall lapse by reason of absence from three successive annual meetings without reason given acceptable to the Council. An excused absence shall not be regarded as an attendance. Membership may be forfeited for reasons deemed sufficient by the Association.

Provided, That in case of removal of an active member to a locality so distant that his attendance at the annual meetings may reasonably be regarded as impossible, the Council may be empowered, should it so desire, to transfer the name of such a member during his absence to the list of emeritus members, without regard to the length of his previous active membership.

There shall be a class of Corresponding Members, not to exceed twenty-five in number, to which distinguished physicians in countries other than the United States and Canada may be elected. The names of those proposed for this membership shall be put in nomination at an annual meeting by two active members, and the Council shall report those nominations which are approved by it to the next annual meeting for election. An affirmative vote of three-quarters of the members present shall be necessary for election. Corresponding Members shall be entitled to take part in the proceedings but not to vote.

ARTICLE III

OFFICERS

The Officers of the Association shall be a President, a Vice-President, a Secretary, a Recorder, a Treasurer—all to be elected annually; five Councillors—one to be elected each year for a term of five years; and a Representative to serve on the Executive Committee of the Congress of American Physicians and Surgeons, to be elected at the annual meeting immediately following the Congress.

ARTICLE IV

DUTY OF OFFICERS

The duties of the President, Vice-President, Secretary, and Treasurer shall be those usually assigned to these officers.

The Recorder shall secure the papers read and also proper notes of the discussions thereon, and shall publish the annual volume of TRANSACTIONS.

The five Councillors shall constitute a Committee of Nomination of Officers, and as such shall present a report at the commencement of the morning session of the last day of the annual meeting, at the conclusion of which session the election shall be held by ballot.

The Representative on the Executive Committee of the Congress shall have full power to represent the Association on all ordinary questions, but must be authorized by the Council of this Association before assenting to the admission of any Society to the Congress.

ARTICLE V

COUNCIL

The officers of the Association shall constitute the Council, of which four members shall constitute a quorum. It shall be the duty of the Council to supervise the affairs of the Association, to make all the arrangements for the annual meeting, to suggest the subject for debate, and to consider the nominations for membership and report on them at the meeting at which they shall be balloted on.

ARTICLE VI

MEETINGS

1. The Association shall hold an annual meeting either in the City of Washington, D. C., or at Atlantic City at a time designated by the Council.

2. The President shall call the meeting to order and open the session by an address not exceeding half an hour in length. In his absence the Vice-President shall preside, and in the absence of both these officers, the senior Councillor.

3. Then shall follow original communications, discussions on subjects of general interest in the department of medicine and pathology, demonstrations in pathology, and miscellaneous business.

4. The President shall have complete charge of, and responsibility for, the program. He shall have power to accept or reject any paper offered. He shall have power to limit the number of papers, and to assign a time limit for each one. Papers shall be presented to the Association only in abstract, and preferably shall not be read from manuscript. They may be published in full in the TRANSACTIONS as also in any other medical publication.

5. The referee, co-referee, and authors of papers shall send abstracts of their papers to the Council for distribution to members previous to the meeting.

ARTICLE VII

QUORUM

Any number of members present at the appointed time of the annual meeting shall constitute a quorum for the transaction of ordinary busi-

ness, but for the election of members, twenty-five shall be necessary for a quorum, and for altering the Constitution fifty members shall be necessary.

ARTICLE VIII

ELECTION OF MEMBERS

Nominations to membership, signed by two members who are not officers of the Association, must be made in writing at an annual meeting of the Association; and within six months of the time of nomination the nominators shall transmit to the Council letters stating the qualifications of the nominee and also a list of his professional positions and publications. The Council shall report the name of each nominee to the members of the Association by letter prior to acting upon the same. The Council shall report those nominations which are approved by it to the next annual meeting for action. The election of new members shall take place on the second day of the meeting. An affirmative vote of three-fourths of the members present shall be necessary for the election of active members, but for the election of honorary members a unanimous vote of those present shall be required.

ARTICLE IX

ANNUAL DUES

The annual dues of the Association shall be such as the Council from time to time may fix, from the payment of which honorary and emeritus members shall be exempt.

ARTICLE X

AMENDMENTS TO THE CONSTITUTION

Amendments to the Constitution must be proposed in writing by five members at an annual meeting, and must be acted upon at the succeeding annual meeting, the notices for which shall contain an announcement of the proposed amendment; and such amendments shall require for their adoption an affirmative vote of three-fourths of those present.

MINUTES

OF THE

THIRTY-SIXTH ANNUAL MEETING

Held at the Hotel Traymore, Atlantic City, N. J., May 10 *and* 11, 1921

The following members were present at the meeting:

Abbott, A. C.
Allen, F. M.
Amoss, H. L.
Anders, J. M.
Austin, J. H.
Barker, L. F.
Bass, C. C.
Billings, Frank
Blumer, George
Boggs, Thomas R.
Brill, Nathan E.
Brooks, Harlow
Brown, Thomas R.
Cannon, W. B.
Capps, Joseph A.
Carter, E. P.
Cecil, R.
Christian, Henry A.
Cohen, Solomon S.
Cohn, Alfred E.
Cole, Rufus I.
Collins, Joseph
Conner, Lewis A.
Dick, G.
Dochez, Alphonse R.
Dock, George
Elliott, C. A.
Emerson, Charles P.
Evans, J. S.
Flexner, Simon
Ford, W. W.
Foster, Nellis B.
Freeman, R. G.
Friedenwald, Julius
Frothingham, C., Jr.
Fulton, F. T.
Funk, E. H.
Fussell, M. Howard
Futcher, Thomas B.
Greene, Charles L.
Griffith, J. P. C.
Hamburger, W. W.
Hamman, Louis V.
Hare, Hobart A.
Hart, T. S.
Heard, J. D.
Herrick, James B.
Hess, A. F.
Hewlett, Albion W.
Hirschfelder, A. D.
Hoover, C. F.
Howard, C. P.
Howard, W. T.
Howland, John
Hun, Henry
Hunt, J. Ramsay
Irons, Ernest E.
Jobling, James W.
Joslin, E. P.
Kober, George M.
Krumbhaar, E. B.
Landis, H. R. M.
Lewis, Paul A.
Libman, E.
Litchfield, L.
Longcope, W. T.
Lord, Frederick T.
McCrae, Thomas
McLester, J. S.
McPhedran, A.
Macht, D. I.
Mallory, F. B.
Means, J. H.
Miller, James A.
Miller, Joseph L.
Minot, G. R.
Morris, Roger S.
Moss, W. L.
Musser, J. H., Jr.
Newburgh, L. N.
Noguchi, Hideyo
Norris, George W.
Ordway, T.
Palmer, W. W.
Park, Wm. H.
Peabody, F. W.
Pepper, O. H. P.
Piersol, George M.
Pratt, Joseph H.
Riesman, David
Robinson, G. Canby
Rowntree, L. G.
Russell, F. F.
Sailer, Joseph
Sewall, Henry
Shattuck, G. C.
Smith, Theobald B.
Sollmann, T. H.
Steiner, Walter R.
Stengel, Alfred
Strong, R. P.
Strouse, S.
Swift, Homer F.
Thayer, W. S.
Tileston, Wilder
Vanderhoof, D.
Wadsworth, A. B.
Walker, J. C.
Warfield, L. M.
White, Franklin W.
White, Wm. Charles
Wolbach, S. B.
Wood, Edward J.
Woodyatt, Rollin T.

Tuesday Morning, May 10

The Association was called to order by President Thayer, who delivered the President's address.

The Treasurer presented his report, and the President appointed Drs. Hoover and Longcope to act as an Auditing Committee.

The Secretary presented the report of the Council, with the following recommendations:

1. That the dues for the coming year be $15 for Active Members who are in practice, $7.50 for Associate Members who are in practice and $5 for both Active and Associate Members in laboratory or full-time clinical work who have no professional income apart from their salaries.

2. The list of nominations, as given elsewhere, for officers and new members.

3. That Dr. Henry Sewall was transferred to the Emeritus list at his own request.

4. That the Meeting in 1922 be held in Washington, D. C., on May 2, 3 and 4.

5. That Dr. W. S. Thayer be reappointed as the Representative of the Association on the National Research Council.

6. That the amendment to the Constitution, creating a class of Corresponding Members, be recommended for adoption with the addition of the words: An affirmative vote of three-quarters of the Members present shall be necessary for election.

The program was then proceeded with:

Dr. L. M. Warfield presented a "Report of a Case of Disseminated Subcutaneous Sporotrichosis with Lung Metastasis," and Dr. Charles P. Emerson presented a paper on "Secondary Mould Infections of the Lung," which were discussed by Drs. L. G. Rowntree, J. B. Herrick, George Blumer, Thomas R. Boggs, S. Solis Cohen and W. S. Thayer, and the discussion closed by Dr. Emerson.

Dr. Simon Flexner and Dr. H. L. Amoss presented a paper "On Experiments in Epidemiology," which was discussed by Dr. Flexner.

Dr. James H. Means and Dr. A. L. Barach presented a paper on "Remarks upon the Symptomatic Treatment of Pneumonia," which was discussed by Drs. George M. Kober, C. F. Hoover and S. Solis-Cohen.

Dr. C. P. Howard and Dr. R. H. Gibson presented a paper on "A Case of Alkaptonuria with a Study of its Metabolism," which was discussed by Drs. T. B. Futcher and W. S. Thayer.

Dr. Frederick T. Lord presented a paper on "The Bearing of the Evolution of the Pneumonic Exudate on Local Chemical Factors

Influencing Recovery and Resolution in Lobar Pneumonia," which was discussed by Dr. T. H. Sollmann, and the discussion closed by Dr. Lord.

Dr. S. Solis-Cohen and Dr. Meyer Solis-Cohen presented a paper on "Observations on the Bactericidal Activity Against Bacillus Diphtheriæ of the Whole, Coagulable Blood of Immunized Animals," which was discussed by Dr. W. H. Park, and the discussion closed by Dr. Cohen.

Dr. W. L. Moss and Dr. G. H. Bigelow presented a paper on "Yaws: an Analysis of 1046 Cases," which was discussed by Drs. E. J. Wood, D. Riesman and J. Collins.

Dr. Richard P. Strong and Dr. W. T. Councilman presented a paper on "Plague-like Infections," which was discussed by Drs. W. W. Ford, H. Noguchi and W. H. Park.

Tuesday Afternoon, May 10

"Discussion on the Causation or Etiology of Infectious and Parasitic Diseases," by Dr. Theobald Smith, Dr. A. R. Dochez and Dr. Hans Zinsser, which was discussed by Drs. S. Flexner and E. Libman.

Tuesday Evening, May 10

Dinner, Hotel Traymore.

Wednesday Morning, May 11

The following Officers and New Members were elected:

President—H. C. Moffitt.

Vice-President—J. B. Herrick.

Secretary—T. McCrae.

Recorder—T. R. Boggs.

Treasurer—J. A. Capps.

Council—A. S. Warthin.

Active Members—R. Cecil, New York; C. W. Edmunds, Ann Arbor; E. F. DuBois, New York; R. Fitz, Rochester; P. A. Lewis, Philadelphia; W. B. MacNider, Chapel Hill; J. H. Means, Boston; G. R. Minot, Boston; H. Zinsser, New York.

Associate Members—W. A. Baetjer, Baltimore; A. M. Chesney, St. Louis; W. W. Herrick, New York; E. S. Kilgore, San Francisco; J. A. Kolmer, Philadelphia; J. P. O'Hare, Boston; P. D. White, Boston; C. S. Williamson, Chicago.

The amendment to the Constitution constituting a class of Corresponding Members was adopted as follows:

There shall be a class of Corresponding Members, not to exceed twenty-five in number, to which distinguished physicians in countries other than the United States and Canada may be elected. The names of those proposed for this membership shall be put in nomination at an annual meeting by two Active Members, and the Council shall report those nominations which are approved by it to the next annual meeting for election. An affirmative vote of three-quarters of the members present shall be necessary for election. Corresponding members shall be entitled to take part in the proceedings but not to vote.

Dr. W. S. Thayer was appointed the Representative of the Council on the National Research Council.

The Auditors reported that the Treasurer's accounts had been audited and found correct.

Dr. S. B. Wolbach presented a paper on "The Pathology and Etiology of Typhus Fever," which was discussed by Drs. A. D. Hirschfelder, E. Libman, R. P. Strong, T. Smith, H. Noguchi, H. F. Swift and N. E. Brill, and the discussion closed by Dr. Wolbach.

Dr. F. B. Mallory, Dr. Frederick Parker, Jr., and Dr. Robert N. Nye presented a paper on "Experimental Pigment Cirrhosis and its Relation to Hemochromatosis," which was discussed by Drs. N. E. Brill, L. G. Rowntree and T. Sollmann, and the discussion closed by Dr. Mallory.

Dr. W. B. Cannon, Dr. J. E. Uridil and Dr. F. R. Griffith presented a paper on "Some Novel Effects Produced by Stimulating the Nerves of the Liver," and Dr. W. B. Cannon and Dr. P. E. Smith presented a paper on "New Evidence of Thyroid Secretion following Stimulation of the Cervical Sympathetic," which was discussed by Dr. H. Sewall and the discussion closed by Dr. Cannon.

Dr. Alfred E. Cohn and Dr. Robert L. Levy presented a paper on "Experimental Studies of the Pharmacology of Quinidin," which was discussed by Drs. A. D. Hirschfelder and G. C. Robinson, and the discussion closed by Dr. Cohn.

Dr. C. F. Hoover presented a paper on "The Relations between Obstruction to Pulmonary Ventilation and the Size of the Lungs," which was discussed by Dr. H. Sewall.

Dr. W. W. Palmer and Dr. W. S. Ladd presented a paper on "Carbohydrate-fat Ratio in Reference to the Production of Ketone-bodied Diabetes," and Dr. R. T. Woodyatt presented a paper on "Objects and Method of Diet Adjustment in Diabetes," which was discussed by Drs. L. N. Newburgh, E. P. Joslin, F. M. Allen, and S. Strouse, and the discussions closed by Drs. Palmer and Woodyatt.

Wednesday Afternoon, May 11

Dr. H. R. M. Landis presented a paper on "Familial Diabetes," which was discussed by Drs. S. S. Cohen, J. M. Anders and F. M. Allen.

Dr. Julius Friedenwald and Dr. George H. Grove presented a paper on "Further Observations on the Blood-sugar Tolerance Test as an Aid in the Diagnosis of Gastro-intestinal Carcinoma."

Dr. James M. Anders presented a paper on "The Relation of Acromegaly to Thyroid Diseases, with a Statistical Study."

Dr. George R. Minot and Dr. L. W. Smith presented a paper on "The Blood in Tetrachlorethane Poisoning."

Dr. Harlow Brooks presented a paper on "A Method for the Study of Early Arterial Changes," which was discussed by Dr. S. B. Wolbach, and the discussion closed by Dr. Brooks.

Dr. E. J. Wood presented a paper on "Further Study of the Estimation of Quantitative Variations in the Vibration Sensation."

Dr. L. G. Rowntree, Dr. E. E. Larsen and Dr. J. F. Wies presented a paper on "Pituitary Extract and Water Balance," which was discussed by Drs. H. Brooks and D. I. Macht, and the discussion closed by Dr. Rowntree.

Dr. John H. Musser, Jr., presented a paper on "The Influence of Iron on Regeneration of the Blood after Anemias."

Dr. Arthur D. Hirschfelder presented a paper on "Applications of Saligenin and Other Aromatic Alcohols and Their Derivatives," which was discussed by Dr. D. I. Macht and the discussion closed by Dr. Hirschfelder.

Dr. David I. Macht and Dr. William Bloom presented a paper on "An Experimental Inquiry into the Cerebral Manifestations of Digitaloids," which was discussed by Dr. A. D. Hirschfelder.

The following papers were read by title: "Manifest Tetany Observed in Association with a Pathological Hyperpnea in a Patient Convalescent from Lethargic Encephalitis," by Dr. Lewellys F. Barker and Dr. Thomas P. Sprunt; "The Adrenals and Thyroid in Experimental Tuberculosis," by Dr. Gerald B. Webb, Dr. G. B. Gilbert and Dr. C. T. Ryder; "A Case of Multiple Polypi of the Stomach," by Dr. A. McPhedran; "The Passage of the Duodenal Tube, Fluoroscopic Observations in One Hundred Consecutive Cases," by Dr. Franklin W. White; "On the Action of Bacterial Culture Products on Phagocytosis," by Dr. Augustus B. Wadsworth; "Experiments on the Etiology of Epidemic Encephalitis," by Dr. E. C. Rosenow; "Symptoms of Appendicitis in Pericarditis," by Dr. M. Howard Fussell; "Further Observations on the Absence of Pancreatic Secretion in Sprue," by Dr. Thomas R. Brown; "A Study of a Group of Symptoms Associated

with Large Thymus in Infants and Children," by Dr. Rowland G. Freeman.

The following amendment to the Constitution was presented: That Section 4, Article VI, be so amended as to read after the sentence "Papers shall be presented, etc.," as follows:

Each member or guest having a paper on the program is under obligation to furnish a copy of his paper or a sufficient abstract thereof to the Recorder for publication in the TRANSACTIONS. The time limit for the receipt of manuscript and for the closure of the TRANSACTIONS to further additions shall be fixed by the Council.

WILLIAM W. FORD,
THOMAS R. BOGGS,
JOSEPH H. PRATT,
HARLOW BROOKS,
ALFRED E. COHN,
LEWELLYS F. BARKER.

A unanimous vote of thanks was extended to Dr. Thayer for the interesting program.

THOMAS MCCRAE,
Secretary.

PRESIDENT'S ADDRESS

By WILLIAM S. THAYER, M.D.
BALTIMORE, MD.

In the year that has passed this Association has lost two of its most distinguished members—Gorgas and Meltzer. This is not the occasion for extended comment on the careers of these two unusual men, but it would be impossible to enter into the activities of the day without a word of affectionate tribute:

Gorgas, Major-General, Surgeon-General of the Army. A native of Alabama; a graduate of the University of the South and of Bellevue Hospital Medical College. For nearly forty years a surgeon in the Medical Corps of the United States Army, for twenty-one years an efficient officer in the discharge of his routine duties, he found himself, in 1901, Chief Sanitary Officer in Havana. Quick to appreciate the significance of the discoveries of the Yellow Fever Commission, he so applied the information they had furnished that in a few months a plague of centuries was banished from the city. And in the years that followed, his conceptions, his initiative and his execution of appropriate prophylactic measures at Panama rendered possible the construction of the canal. These two examples of the intelligent, carefully studied, accurate application of definite biological information to the problems of public sanitation in the tropics are models for all time. From this moment the counsel and advice of Gorgas were universally sought. Surgeon-General during the late war, with the rank of Major-General, Permanent Director of the International Health Board of the Rockefeller Foundation, honoured throughout the world by universities, scientific societies, foreign governments, he died in London while on an important mission to Africa.

Meltzer, physiologist. Born in Russia; educated in Germany; a practitioner of medicine who, throughout his life, combined with his daily duties as a doctor, scientific studies of importance such as to place him among the leading physiologists of his day. More than any other man in American medicine, Meltzer has stood out as the figure proclaiming to the world that the life blood of the medical art springs from its connections with the underlying and associated sciences. The keynote of his career, which never varied throughout his long life, was the message: "To do your work in the world as a doctor you must be a student. To be a worthy associate of other students you must be an observer, an investigator, a contributor to the common store of knowledge." To the day of his death, and he was nearly eighty, he exemplified that which he taught.

These two men, so different in many respects, met here on common ground—a circumstance which illustrates well the essential character of this Association, which consists of a limited group of men with a wide variety of special interests—clinical, physiological, pathological, bacteriological, chemical, physical—who are joined together by the common bond of devotion to the cause of the advancement of the science and art of medicine.

In a larger way it represents the elements which go today to make up the medical clinic, which, for the elucidation of the daily problems concerning the nature and treatment of disease, demands not only its wards but its laboratories—pathological, physiological, biological, chemical—with their workers trained especially in special fields. Differentiation of function is as vital in a medical clinic or in medical practice as it is among the organs that go to make up the human mechanism; but, as in the human mechanism so in the clinic or in a body like ours, close association, community of interest, coöperation, are vital alike to the part and to the whole.

If this Association shall continue to fulfil the hopes of its founders it will always consist of just such a group of students, representing many special interests, but united in the common study of physiology, pathology and therapy—medicine in its broadest sense.

It is a remarkable evidence of the foresight of that group of men who conceived this society that there is nothing in its constitution—

which is a model of simplicity and brevity—which defines the qualifications for membership; nothing which demands that members shall be possessed even of a medical degree. From its very nature this body can never contain more than a small part of those whose qualifications might justify membership. In this respect it has always had many of the aspects of a club, and although this restriction of membership is, as I can testify from long experience, a sad burden to the Council which has yearly to select candidates for membership from a list constituted largely if not entirely of desirable names, yet it would appear to be necessary for the preservation of the intimate character of the organization.

At the opening session of the Association of American Physicians in 1886, the first President, a wise and foreseeing man, made a remarkable address—remarkable for its force, its comprehensiveness and its brevity. Despite his protest, it sets forth the objects for which we stand with remarkable directness. It may not be amiss after thirty-five years, to repeat his words:

"Gentlemen: At the inauguration of a new medical association it would seem proper for the presiding officer to say some words concerning the objects of the association and the best methods of carrying those objects into effect, and it was my intention to do this; but when I considered the character and attainments of those whom I was about to address, such a plan seemed to me to be unnecessary and even impertinent.

"We all of us know why we are assembled here today. It is because we want an association in which there shall be no medical politics and no medical ethics; an association in which no one will care who are the officers and who are not; in which we shall not ask from what part of the country a man comes, but whether he has done good work and will do more; whether he has something to say worth hearing and can say it. We want an association composed of members, each one of whom is able to contribute something real to the common stock of knowledge, and where he who reads such a contribution feels sure of a discriminating audience.

"We are all busy men, working men, ambitious men. We all like to give instruction and to gain reputation, and both of these we can do in the societies already existing. But we also want a

society in which we can *learn* something. And this, I take it, is the real object of the enterprise which we inaugurate today—to form an Association of Physicians and Pathologists to which we may come, year after year, with the well-founded hope that at each meeting we shall find something to *learn*.

"Without further preface, I shall ask your attention to a subject of much practical importance."

Gentlemen, without further preface let us proceed to the discussions of the day.

REPORT OF A CASE OF DISSEMINATED GUMMATOUS SPOROTRICHOSIS WITH LUNG METASTASIS

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Diseases due to certain varieties of fungi are claiming more and more attention. Scattered through the literature are many case reports and careful bacteriologic investigations of actinomycosis, blastomycosis, streptothricosis, sporotrichosis, etc. Some of the fungus diseases are primarily pathogenic for animals and only now and then produce disease in man when conditions are favorable. Many of the forms of fungi occasionally pathogenic for man are in their natural state saprophytes on dead vegetables, bark of trees, hay, etc. Gradually the various forms have been differentiated one from the other until now there are quite a number of well-recognized mycelium-bearing fungi which may cause even fatal disease in man. Among these Gougerot gives an important place to the sporotrichum. In an address delivered in 1913 he gives six reasons why sporotrichosis should claim attention:

1. Because of its extreme frequency (in France at least) and of its universal distribution: There is no part of the civilized globe which has not had its case or cases. In France over 200 cases were reported in the literature from 1906 to the end of 1910. Since then no one reports his cases unless there is something quite unusual about them.

2. Because of the practical importance from both prognostic and therapeutic standpoints of a correct diagnosis: He cites a number of cases in which the patients were given several months to a year of rest-cure under the diagnosis of pulmonary tuberculosis. In one case reported a man had had a hip-joint amputation, then a forearm

amputation was advised but refused. Diagnosis was corrected and the patient was well in six weeks. One case had severe pharyngeal ulcerations which were thought to be syphilitic. Mercury had been given, with no relief. Diagnosis was finally corrected and the patient was soon cured.

3. Because of the polymorphism of the disease there is no tissue exempt from its ravages, including bone.

He makes three main groups with subgroups:

I. Disseminated sporotrichosis.

(*a*) Disseminated gummatous non-ulcerating variety of the skin.

(*b*) Subcutaneous ulcerating tuberculoid gummata.

(*c*) Ulcerating syphiloid gummata.

(*d*) Polymorphous sporotrichosis.

II. Localized sporotrichosis.

III. Extracutaneous sporotrichosis—osseous, ocular, visceral, etc.

4. Because of the great ease in diagnosis one does not need a laboratory nor does he need to be an expert bacteriologist. He does need, however, tubes of glucose-agar. By inoculating a tube with some of the purulent material in the ulcers or abscesses and placing it aside at room temperature for four to ten days the macroscopic appearance is characteristic.

5. Because of the almost certain healing of even the most extensive lesions by the internal administration of KI, necessitating no loss of time by the patient from his daily occupation.

6. Because in the intensive study of the disease a new impetus was given to the study of mycotic diseases in general.

The honor of calling attention to the pathogenicity of the sporotrichum goes to B. F. Schenck, of Johns Hopkins, who in 1898 published an article in the *Bulletin of the Johns Hopkins Hospital* on "Refractory Subcutaneous Abscesses Caused by a Fungus Possibly Related to the Sporotricha." He isolated and grew out the organism which has since been known by the name of S. schenckii. In 1900 Hektoen and Perkins published a long article on the sporothrix. The first case published in any other country was that by de Buermann in 1903. Four years elapsed with the report of only four cases seen by de Buermann and Gougerot, then the *Bulletin et mémoires*

de société médicale des hôpitaux de Paris was full of reports. From that time cases have been reported from various countries and from most of the states in the United States. Hyde and Davis, of Chicago, reported on the disease in 1910.

In 1912 Ruediger was able to collect 57 cases from the American literature. Only 24 of these were culturally proved cases. There was one brief report of a probable case from northern Wisconsin. An analysis of the cases revealed that the largest number from any single state was from North Dakota and that most of the cases occurred along the valley of the Missouri River. Sutton had previously reported a case, and within a year 11 cases had been reported from the Missouri Valley. Haynes and Cheney reported the first case in West Virginia in 1914.

Blaisdell, in 1917, reported the first case in New England. None have been reported since, although cases must have been seen since then.

The sporothrix has been found in every tissue of the body. The common type in this country is the localized lesion. Following an injury a nodule appears, often on the tip of a finger, ulcerates and discharges pus. Healing is very slow or does not occur. Within a short time a nodule forms along the lymph channels which drain the part and the lymph vessels become cord-like. Farther up another nodule forms connected with the first by an indurated line of tissue slightly or not at all reddened. Others may form along the same line so that a chain of several separate nodules may be seen along the arm or leg. They may ulcerate and cicatrize or become soft and break down with ulceration. Frequently if the nodules are close together the tissues are undermined and a probe may be passed in the subcutaneous tissue from one ulcer to another. As a rule there is not much tendency to spontaneous healing. The ulcers may last for weeks or months. The course is usually very slow. There are no disseminated gummatous lesions reported in this country.

In France especially there have been many widespread subcutaneous nodules and ulcerating forms reported. However widespread the lesions they always begin as a firm, usually painless but sometimes very tender nodule in the subcutaneous tissue. In

the disseminated form every nodule is separate and no cord-like tissue links it to its nearest fellow. After growing to a variable size the skin over the nodule becomes purplish, especially at the apex, and quite thinned, and fluctuation is felt. The nodule breaks, releasing a sanguinopurulent material. There is little or no zone of inflammation around the base of the nodule. The resulting ulcer has a punched-out appearance resembling so much a syphilitic ulcer that many errors in diagnosis have been made. The ulcer may show signs of healing, the base becomes clean and healthy granulations appear while the epithelium grows in from the periphery. While some may be healing other nodules and ulcers are forming.

Although de Buermann and Gougerot stated in 1907 that the regional lymphatic glands do not enlarge, this is not altogether true. They do not show the same tendency to become involved as do the glands draining tuberculous or syphilitic skin lesions, but they do enlarge and even become tender in many cases.

De Buermann and Gougerot in 1907 reported the thirteenth case. This was an ascending gummatous lymphangitis located on the arm of a man fifty-seven years old. It was the first case of the kind reported in France. A period of eighteen months elapsed between the appearance of the first and second abscess. In this case the axillary glands were not enlarged. That the course of the lymphangitis is not always as described above is shown by the case of Dominici and Rubens. The initial lesion was the result of a finger cut. Later there were numerous scattered subcutaneous gummata on the arm without lymphangitis and without swelling of the lymph glands.

De Buermann and Gougerot saw a sporotrichal chancre of the forehead. Alvin and Vallin report a case of a large abscess and fistula in the brow of a woman aged fifty-one.

Danlos and Blanc report a case of palpebral sporotrichosis.

Wilder reports conjunctival ulceration, culturally proved, in a laboratory assistant who was working with cultures of the sporothrix, and frequently capillary tubes were broken within ten inches of his eye. Morax reports a most unusual case of uveal sporotrichosis with secondary episcleral lesions and absence of all other discoverable sporotrichal localizations.

De Buermann and Gougerot reported the first case of sporotrichosis of the pharyngeal mucous membrane. The organism was grown from the ulcer and also from the apparently intact mucous membrane.

Letulle and Klebré in the course of a report on a case of sporotrichosis of the skin, mouth, pharynx, larynx and trachea, in which the whole posterior buccal cavity was a shallow ulcer, say that the sporotrichum buermannii can live an indefinite time in a saprophytic state.

Monier-Vinard and Gaucher, in 1907, reported the first case where the sporotrichum was grown from the sputum.

Widal and Weill reported, in 1908, a case of disseminated sporotrichosis with periosteal lesions in which the sporotrichum was recovered from the circulating blood. Widal and Abrami also showed at this time that the spores of the sporotrichum were agglutinated by the blood of patients affected with the disease in dilutions of 1 to 800. The blood serum of a patient with actinomycosis agglutinated the sporotrichum in dilutions of 1 to 150. Complement-fixation with the spores as antigen has also been described.

The disease usually begins as a local lesion at the site of some injury. The fact that the fungus lives as a saprophyte on all kinds of vegetables and dead matter and bark of trees makes it strange that the infections among humans are so few. Some curious sources of infection have been reported. Thus, Sutton reports a case due to a hen bite, Rouslacroix to a parrot bite and others to bites of horses. Olson reported a case of the lymphangitic form in a man from North Dakota who had killed and handled many gophers, many of which had sores similar to those on the patient's hand. Many cases began from the crushing of a finger-tip from cuts while in hay fields. Several cases are reported by the French of disseminated type in which no portal of entry could be found. The patients were vegetable venders and the organism was found on the vegetables. Meyer thinks that flies may act as passive carriers. In horses, dogs and rats in this country sporotrichosis is found spontaneously. Davis found that a small epidemic among horses on a farm in North Dakota which was thought to be farcy was sporotrichosis. The disease was also found among horses in Pennsylvania.

Bovines do not seem to be susceptible to the disease, but the hides may act as transmitters of spores (Meyer).

All the American cases, with few exceptions, which I have been able to find have been of the lymphangitic type. A case reported by Ravogli, in 1913, had a huge tuberculoid ulceration around the elbow. Hyde and Davis's case was apparently the gummatous form. Le Blanc has recently briefly recorded two cases of what had been thought to be pulmonary tuberculosis. He saw a small nodule on the larynx of each one and claimed that he found spores of sporotrichum in the sputum. There were no skin lesions.

Dominguez, of Cuba, reported, in 1914, a case in a tobacco-leaf tester who was ill with fever and lung findings of pneumonia. His illness was prolonged and no tubercle bacilli could be found in the sputum. By *x*-ray plate a shadow was found in the lung. A fungus lesion was then suspected and sporotrichum was grown from the sputum. Later an abscess of the middle ear developed from which sporotrichum was grown. An extradural abscess was operated and drained, but culture was negative for the fungus. The author found sporotrichum on the tobacco leaves.

Besides these lesions cases are reported by the French of genital lesions and muscle and bone lesions, and Councilman grew a sporotrichum from the fluid of a swollen and inflamed knee-joint.

The case here reported is the only one of its kind recorded up to the present in the United States. The complete report of the bacteriologic, experimental and immunologic studies will be pubpublished later by my assistant, Arthur Margot.

The unusual character of the lesions with localization in the lung is the excuse for bringing a single case report before this Association.

Case History.—The case is that of a young negro, twenty-three years old, admitted to the Milwaukee County Hospital, March 8, 1920, who had followed the race horses as a stable boy in the southern and western circuits. He was born in Arkansas. Except for his journeys from New Orleans to California with the horses and a recent trip to Detroit he has always lived in the southern part of the country. He came to Milwaukee direct from Detroit and was in the city for five months before he was taken ill.

The family history is negative. As a boy he had measles, whooping-cough and mumps. When nineteen he had swollen and painful joints lasting a month, but was not confined to bed. At the age of fifteen he had both gonorrhea and an "eating chancre," without suppuration of the inguinal glands. He does not drink but smokes cigarettes to excess. He dated his present illness as far back as June, 1919, when he was ill with fever and chills, but was not confined to bed. Ever since then he has felt weak. About two weeks before admission a swelling appeared on the top of the right foot. There were no ulcerations of the toes, but the toes felt "sprung." This swelling was painful, at first hard, then later soft. It broke in about a week and yellow matter came out. It then began to heal. In the meantime nodules appeared on the top of the left foot, on both upper arms, posterior surfaces, both cheek bones and forehead. He became so weak that he entered the hospital.

Physical examination revealed a somewhat emaciated negro of medium height. On the left side of the forehead at the hair line was a soft painful swelling. There was slight dark discoloration of the skin of the nodule, and it was sharply demarcated from the surrounding skin. It measured 2 cm. in diameter and was raised about 1.5 cm. There was a rough bony edge encircling the base, as if the bone had been eroded. Behind the right ear was a tender superficial ulceration covered by a thin crust. The face was distorted by a swelling in the skin over each malar bone. One was somewhat softer than the other. On the exterior surfaces of both lower arms there were nodules, the largest measuring 3 cm. in diameter. All nodules were tender, some were discolored and others not discolored. All showed more or less fluctuation. They appeared to be in the subcutaneous tissue. On the dorsum of the right foot there were three superficial ulcers, two of them almost healed; one measured 3 x 4 cm. and had a healthy granular base and a punched-out appearance.

There were moist rales at the bases of both lungs, more numerous on the left side. The skin of the left posterior apex was very sensitive to palpation. The percussion note was slightly impaired and the breath sounds were a trifle harsh. No rales were heard. Over the whole left side the respiratory excursion was slightly limited and

the tactile fremitus, percussion note and breath sounds were not as distinct as on the right side.

The heart was not enlarged; there were no murmurs; the rate was 90 to the minute; the pulse was soft and compressible.

The abdomen was retracted; no masses were felt; the liver and spleen were normal in size; the reflexes were normal.

There was irregular fever as high as 103° F. throughout the illness. The urine was normal at first, later contained a slight amount of albumin and shortly before death was full of bile pigments. The leukocytes were 17,000, the Wassermann reaction was ++++.

As soon as he was seen by me he was thought to have an infection with some one of the fungus diseases. A soft nodule was aspirated under aseptic precautions and the yellow blood-stained pus was examined fresh and cultured. A mycelial network was found which on culture proved to be sporothrix.

Suspecting that there might be some lesion in the lung an *x*-ray plate was made and the sputum examined. Nothing of interest was found in the sputum, which was scanty.

The *x*-ray plate showed a clear right apex and slightly hazy left apex. There was a large somewhat circular shadow extending from the right hilum, about 6 cm. in diameter. Radiating from the edge of this were fine linear markings extending into the first interspace and outward to the edge of the outer zone. Along the left heart border was some fine spotting. The left hilum was also increased in size.

In spite of large doses of potassium iodide and of arsphenamin new nodules developed at various places on the arms and legs while those on the face were gradually healing. Over the outer surfaces of both thighs very large undermined nodules developed. Cultures made from the contents of many nodules invariably showed the same sporothrix.

On April 1 it was noted that on the inner side of the middle of the left thigh was an elongated hard nodule from which a cord-like induration extended a short distance in both directions. Shortly after this he developed intense jaundice. One nodule over the left malar bone had increased in size; the skin was thin and bluish, but there was no elevation of temperature. Ten cc of a yellow purulent blood-stained fluid was aspirated from this nodule.

The patient gradually grew worse. Many nodules were open and were draining freely. The temperature was always elevated. He died on May 23, 1920.

It was not possible to autopsy the body until June 2, 1920, ten days after death. The report of Dr. Margot is as follows:

The body is that of a large-framed but extremely emaciated negro male. The legs and arms are swathed in bandages. Upon removal there are seen huge ulcerating surfaces with undermined discolored skin, one enormous cavity occupying a large part of the outer surface of the left thigh. At the scalp margin on the brow are two punched-out ulcerations. In the skin over the right malar bone is a fluctuating tumor 4 cm. in diameter and 2 cm. above the skin level. Over the left malar bone is a similar but larger nodule. The conjunctivæ have an icteric tint. The pupils are equal but rather large. Rigor mortis is present; postmortem lividity is marked. There is no edema.

Thorax. On removing the sternum the left lung lies entirely free from adhesions but shows a slight hypostatic congestion in the posterior portion of the lower lobe. There is an area of congestion on the posterior surface of the lower lobe of the right lung. A nodule about 1.5 cm. in diameter is found in the upper right lobe having the characteristics of those found in the skin. The mediastinum contains no enlarged glands.

Heart. The pericardium and heart reveal no abnormalities except for probable softening of the muscle.

Abdomen. The abdominal fat is scanty and has a yellow color. The peritoneal surfaces are smooth and glistening. No free fluid is found. The spleen is slightly enlarged; the surface is smooth; color dark; consistency irregular, soft and firm. On section there are dark red dry areas which are firmer than the rest and extend deep into the spleen. The Malpighian bodies are pretty well seen throughout the cut surface.

Liver. The liver extends 4 cm. below the costal border. It is mottled and the external surface is smooth. It is firm. On section it drips blood. The cut surface is somewhat glistening and the cut edges roll over and the markings are indistinct.

Kidney. The kidneys are slightly increased in size and are firm. The capsule strips readily from both. Typical cloudy swelling markings are seen with swollen cortex.

Gall-bladder. The gall-bladder is distended and contains about 45 cc of purulent exudate and bile. The wall is thickened and edematous. No stones are found.

Pancreas. The pancreas appears not to be affected.

Microscopic Examination of Tissues. Owing to the fact that the body was ten days in the morgue before autopsy the tissues were more or less autolyzed. However it was possible to obtain good specimens of the lung. Sections of the nodule in the right lung are made up of connective tissue in which are small round cells, scattered polynuclear leukocytes, plasma cells and large mononucleated cells probably from the alveolar epithelium. The lung structure is completely obliterated. Many giant cells are present. These have irregular shapes, the protoplasm stains a uniform pink with the eosin and the many apparently vesicular nuclei are arranged along the periphery of the cells, at times almost encircling the cell. The resemblance to the giant cells of tuberculous tissue is very close. There are lacking the caseous areas and the tubercle arrangement. There are many distended capillaries throughout.

The kidney sections show marked cloudy swelling with detritus in the collecting tubules. Many of Bowman's capsules are unusually thickened.

The liver sections show cloudy swelling and congestion of the central veins with atrophy of the liver cells for about one-third to one-fourth of the lobule.

No special changes are seen in the spleen or in a lymph gland at the hilum of the right lung.

Sections from the edge of several of the skin nodules show a connective-tissue stroma infiltrated with leukocytes, round cells and many giant cells much like those found in tuberculosis. Very few eosinophils were seen. Early stages of the nodules are made up of embryologic connective tissue infiltrated with many plasma cells and some polynuclear cells. As the nodule softens the leukocytes increase in number. No proliferation of capillaries is seen. Giant cells are found. It is often quite impossible in certain areas to differentiate these lesions from those of tuberculosis.

We grew the organism from the pus of several nodules on ordinary nutrient agar-agar in the incubator and at room temperature. The best mediums were potato and Sabouraud's glucose peptone agar. At room temperature the growth is slow. In the incubator on agar at the end of twenty-four hours small circular colonies of silky-white appearance are seen and a dense branching, silken-whitish mycelium spreads over the surface. In tubes this penetrates vertically for a distance of 5 to 10 mm. into the depths of the medium. In addition to this adherent layer of interlacing filaments an abundant aërial woolly mass of filaments appears within forty-eight hours. Radiations of short filaments from the center of the growth on the agar surface are found from which fruiting hyphæ arise, at the free ends of which are sporangia. The columella, seen after the discharge of spores, projects in a spherical body from the funnel-shaped expanding end of the supporting hyphæ. The spores are conical in shape; they are chlamydospores (Davis) and measure about 4.5 micra in length, although the range is from 4 to 5 micra.

After four or five weeks the growth on the agar or potato appears dry and the color changes from white to mouse gray on to dark brown. De Buermann, who described the first case in France five years after Schenck had published his report, has given his name to the organism, and possibly on account of his many publications the name of the sporotrichum is more widely known under the name Sporotrichum buermanni, than under its correct name, Sporotrichum schenckii. Priority, however, in this case is undisputed and the organism should be known by its original name.

It would appear that de Buermann's organism and our organism are slightly different in their pathogenicity from the one described by Schenck and the ones described by Hektoen and Perkins, Hyde and Davis and others in this country. The difference lies in the ability of the first group to produce lung lesions in albino rats, whereas no lung lesions were ever produced by any of the previously described American sporotricha. Culturally there is no essential difference. Since Davis reported that the Sporotrichum schenckii produced chlamydospores, that distinction of the two species no longer served to differentiate them.

It is interesting that our case, the only one reported in this country which is similar to the gummatous cases reported in France, should be caused by a sporotrichum different in pathogenicity from all the reported American cases, but like that reported by the French investigators. We procured cultures from various parts of the country and injected them into rats. Our culture was the only one which produced lung lesions.

The spores alone develop and germinate in the organs of animals (Margot). An emulsion of spores was injected intravenously into rabbits, albino rats and guinea-pigs. Within ten to twelve days the animals developed cachexia, and if not killed died from the injection. Autopsy showed nodules scattered in the liver, kidney, spleen and brain. The nodules were subcapsular, deep, isolated and conglomerate. Cultures from the organs developed typical mycelium.

Only in the albino rats were lung lesions produced. Inoculation by any route invariably produced nodules in the lungs. The histologic appearance of the nodules was similar always, only varying in the proportion of giant cells and other formed elements. No spores were definitely found in the giant cells.

Our organism was agglutinated by the patient's blood (spore-agglutination) in dilution of 1 to 150. Control sera gave no agglutination.

By using an emulsion of crushed spores as antigen complement-fixation was obtained. Unfortunately we neglected to draw blood from the living patient for culture. The result would have been interesting but of no value in diagnosis.

Summary. A case of disseminated gummatous sporotrichosis in a young colored man is reported. There was a nodule in the right lung, suspected on physical examination, seen in the *x*-ray plate and confirmed at autopsy culturally and histologically. This is the first case of its kind reported in American literature. The sporotricha reported from American cases and some reported from French cases differ in the pathogenicity and in the ability to produce lung lesions in albino rats.

Attention is again called to sporotrichosis as a generalized disease and to the probability of cases of lung sporotrichosis which are unrecognized.

BIBLIOGRAPHY

Schenck, B. F.: On Refractory Subcutaneous Abscesses Caused by a Fungus Possibly Related to the Sporotricha, Bull. Johns Hopkins Hosp., 1898, ix, 286.

Hyde and Davis: Sporotrichosis in Man with Incidental Consideration of its Relation to Mycotic Lymphangitis in Horses, Jour. Cutan. Dis., July, 1910, xxviii, 321. (Complete bibliography to date, including the articles published during 1907 and 1908, in Bull. et mém. Soc. méd. d. hôp. de Paris.)

Adamson, H. G.: A Résumé of the Literature Relating to Sporotrichal Infections of the Skin, British Jour. Dermatol., 1908, xx, 296. (All literature to date.)

Ruediger, G. F.: Sporotrichosis in the United States, Jour. Infect. Dis., 1912, xi, 193.

Hamburger, W. W.: Sporotrichosis, Jour. Am. Med. Assn., November 2, 1912, lix, 1590.

Olson: A Case of Sporotrichosis, Ibid., September 21, 1912, lix, 941.

Ravogli, A.: Report of a Case of Sporotrichosis of the Tuberculoid Type, Lancet Clinic, Cincinnati, 1913, cx, 112.

Dominguez, F.: Sporotrichosis with Multiple Localizations, Med. Record, April 4, 1914, lxxxv, 608.

Gougerot: Sporotrichosis, Jour. State Med., London, December, 1913, xxi, 705–768.

Hamburger, W. W.: Sporotrichosis in the United States, Illinois Med. Jour., February, 1914, vol. xxv.

Quém, E.: Sporotrichosis of Breast, Rev. de Chir., Paris, 1914, xxxiv, 553–720.

Wilder, W. H., and McCullough, C. P.: Sporotrichosis of the Eye, Jour. Am. Med. Assn., 1914, lxii, 1133.

Sutton, R. L.: Sporotrichosis in the Mississippi Basin, Ibid., 1914, lxiii, 1153.

Davis, D. J.: Formation of Chlamydospores in Sporothrix Schenckii, Jour. Infect. Dis., November, 1914, vol. xv.

Morax: Uveal Sporotrichosis with Secondary Episcleral Sporotrichosis; Absence of All Other Discoverable Sporotrichal Localization, Ann. d'ocul., Paris, 1914, clii, 273.

Davis, B. F.: Report of a Case of Sporotrichosis, Surg., Gynec. and Obst., 1914, xix, 490.

Haynes and Cherry: Case of Sporotrichosis, First Case in West Virginia, West Virginia Med. Jour., 1914–15, ix, 303.

Blaisdell, J. Harper: A Case of Sporotrichosis, First Case in New England, Jour. Cutan. Dis., 1917, xxxv, 452.

Meyer, K. F.: The Relation of Animal to Human Sporotrichosis, Jour. Am. Med. Assn., 1915, lxv, 579.

Alvin, H., and Vallin, H. G.: Sporotrichosis, Lyon méd., October, 1920, cxxix, 859.

Wohl, M. G.: Report of a Case, Nebraska State Med. Jour., December, 1920, v, 355.

Le Blanc, F.: Pulmonary Infection by Sporothrix of Schenck, Illinois Med. Jour., December, 1920, xxxviii, 516.

Brainos, A.: Sporotrichosis of the Genital Organs, Paris méd., March, 1920, x, 247.

PENICILLIUM LUNG INFECTION

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Of a group of pneumonomycoses which during the past few years it has been our privilege to study, two cases have been so interesting that we take the liberty to bring them to your attention. These two patients are suffering from mixed pulmonary infection, and one of the infecting organisms in each case is a penicillium mould.

In nearly all medical literature the penicillium moulds are stated to be "merely saprophytes" in a manner so emphatic that it seems almost a misdemeanor to accuse them of any pathogenicity whatever. Naturally the prejudice even of one well read in the literature of this subject would tempt him at the onset to conclude that the moulds we found are merely contaminations.

The first patient, "W," a man, aged fifty-three years, we found one morning six years ago in the ward. He said that for seven years he had been considered a consumptive, and yet the symptoms he related did not at all suggest pulmonary tuberculosis.

No. 1031. J. W., aged forty-seven years, male, white, admitted to the Robert W. Long Hospital May 2, 1915; discharged June 5, 1915. The patient complained of "throat trouble, choking and shortness of breath."

Family History. His father, who had been "doctored for his heart," was found dead in bed. His mother died at the age of fifty-five of "complications." One brother, two years old, died of whooping-cough. Of four sisters, one is living and well, one died two years old of "spasms," one fifteen years old of typhoid fever and one twenty years old of "heart trouble." No relative has had tuberculosis. The patient is married and has four healthy children.

Past History. The patient "had the usual diseases of childhood," with good recovery from each, and typhoid fever when twenty years old. He denies chancre and all venereal infections.

The right leg shows the scar of a large tibial ulcer on its outer anterior surface and the left an area of brown pigmentation over the corresponding area. He says he had had sharp, tearing pains in his legs during sleep. He says he was "raised in a grist-mill," where he worked as a miller for years until he was twenty-two years old; then he worked in a butter-tub factory, running a three-gang saw. When the present illness began, seven years ago, he was driving a milk-wagon. He has not worked at all for about seven years. In 1911 he weighed 119 pounds.

Present Illness. His present illness began seven years ago as a chronic cough. At that time he began to have paroxysms of coughing, which commenced with a tickling in the throat and a choking sensation. The mouth then would fill with mucus, which he would expectorate for from five to ten minutes. This sometimes contained "slugs." The coughing was aggravated by stooping over or lying down. These attacks came on irregularly, at no particular time of day, but especially after exertion. Some of the mucus seemed to come down from the nose and to accumulate in the throat. This was noticed particularly when the head was thrown backward. These paroxysms soon became smothering spells, often so severe that he had to rush out of doors to get his breath. Some of these smothering spells could be predicted from four to six hours in advance by the coughing, and were so severe that they required morphin for their relief. He said that about half of his time was taken up with such spells. Because of them he had for seven years not slept in bed, but in a chair. About four years ago his trouble was much worse and was accompanied by high fever and night-sweats. The patient had become so dizzy that he could not ride in the cars or walk fast. He became very short of breath when he climbed up stairs.

His sputum has never been blood-streaked. It had been examined ten or twelve times for tubercle bacilli, but always with negative results. The patient belongs to the Woodmen of the World. About five years ago he applied for admission to their sanitarium in Colorado for tuberculous patients, but was refused "because he was too advanced a case of consumption." Later he applied a second time and was refused "because he was too well." (He explained this on the ground that he had taken a large dose of morphin just before this second examination.) In 1910 he went to Petosky for "asthma," but his symptoms were much worse there.

Physical Examination, May 5, 1915. The patient was sitting in a chair beside his bed, breathing heavily. He was cyanotic and apparently orthopneic. He coughed frequently—a dry, brassy cough. His dyspnea was quite different from that of circulatory cases since if at the time that he seemed to suffer acutely he took one long breath or changed his position he became at once comfortable.

He was poorly nourished. The skin of the entire body was slightly pigmented. It was warm and moist. There was an area, irregular in outline, of brown pigmentation about the size of a dollar over the second left costal cartilage.

There was some fulness of the lower lids. No exophthalmos. The conjunctivæ and scleræ were much injected. The pupils of his eyes reacted normally to light and to accommodation. There was no Joffroy's, no v. Graefe's and no Stelwag's signs. Internal convergence was poor.

The tongue was clean. The teeth were loosened by an extreme grade of pyorrhea. The anterior fauces were very red, the tonsils were flat and with some of their crypts open. Chronic pharyngitis was present. The nasal septum was deviated to the left side. The vocal cords were negative; there was definite nasopharyngitis and laryngitis. The thyroid gland was not enlarged. A few lymph glands were palpable in the right posterior triangle of the neck. The arterial and venous pulsations of the vessels of the neck were not unusual.

The left shoulder was higher than the right. The suprasternal and supraclavicular fossæ were very deep. The chest showed in its shape the habitual stoop of the patient. It was rather flattened in front, especially over the right upper front, yet the sternum was prominent. The ribs were rather too horizontal. The scapulæ tended to flare. Its anteroposterior diameter was not much increased. The respiratory movements were quite limited, the circumference on inspiration being 83.3 cm. and on expiration 82 cm. This limitation of expansion seemed in large part due to the patient's intratracheal difficulty. The expansion over the fronts was poor, that of the right side especially so. Tactile fremitus was increased over both sides from apex to base. The note on percussion over both fronts was definitely impaired, almost flat, especially on the right side over the infraclavicular fossa. On auscultation the breath sounds were harsh on inspiration and prolonged and wheezing on expiration. Musical and moist rales were heard, especially during deep inspiration, over both upper lobes. Vocal fremitus was increased (pectoriloquy). Over both backs and axillæ from apex to base there was impaired resonance on percussion, increased tactile and vocal fremitus (this is more marked on the right side) and breath sounds which were harsh and wheezing and accompanied by musical and moist rales, which were heard everywhere, but in increasing number toward the base.

The pulse was 64 to the minute, regular in force and rhythm. The radial artery did not feel particularly thickened. The systolic blood-pressure was 108 mm. and the diastolic 74 mm. Hg. The point of maximum impulse was indefinitely made out in the fifth interspace near the nipple. The deep cardiac dulness extended 6 cm. to the right in the third and 3 cm. to the left of the nipple line in the fifth interspace. The sounds were distant and faint, the first accompanied by a faint systolic blow, heard

down the left sternal border and at the apex. The pulmonic second was accentuated.

The abdomen was negative. There was cyanosis and slight clubbing of the finger-tips. The nails were longitudinally striated and brittle. The epitrochlear lymph glands were not palpable, but the axillary glands were slightly enlarged. The skin over the ankles pitted slightly on pressure. The knee-kicks and other deep reflexes were normal.

The temperature rose not higher than 99.2° during the first three days (May 2, 3 and 4) that the patient was in the hospital. After that (May 5 to June 5, 1915) it did not rise above 98.5°.

The urine was straw-colored; its specific gravity in the morning varied from 1015 to 1020; it was acid in reaction and contained no sugar and only a trace of albumin. The sediment contained epithelial cells, pus cells and coarsely and finely granular casts.

The sputum consisted of a tenacious ropy mucus, light gray in color, which was streaked with threads and masses of pus. It was negative for Bacillus tuberculosis on May 4, 5, 7 and 11, 1915. It contained staphylococci, pneumococci, streptococci and the spores and mycelial threads of a mould.

Sputum Cultures. The mouth and teeth were thoroughly cleansed at bedtime and again on rising the following morning. The sputum he raised that morning was collected in a sterile beaker, washed at once and cultures made. A penicillium mould grew on each plate. Controls were negative.

Wassermann test, May 3, 1915, + + (50 per cent).

Blood culture taken May 6, 1915, was reported May 24, 1915, positive for a long chain streptococcus.

Treatment. Potassium iodide in increasing doses.

Second Admission. No. 1921. Admitted November 9, 1915, for demonstration to the medical class and discharged November 11, 1915. Temperature normal.

Third Admission. No. 6331, April 28, 1918. Discharged April 29, 1918. Patient said that one month more would complete the second year he had been working as night watchman. He said that if he moved a little too fast he would have a coughing spell, but now without sputum. He said that he could not lie down in bed at night, but sits up in a chair or piles the pillows up against the headboard and sits on the bed. It is not the shortness of breath so much as a tickling in the throat and a cough, which even made the tears run down his cheeks, which has prevented him from lying down. In fact there was no definite shortness of breath when he does lie down. If he moved too fast when working his breath would seem short, but if he draws one long breath he is all right. He then had no pain across the chest. His weight at that time was 135 pounds. He had had no chills or chilly feelings. His appetite in general was good; there was no nausea or vomiting and the bowels were regular.

Physical Examination. (We give only the important points of difference from the former examination.) The patient certainly had gained in weight. The left shoulder was higher than the right and expansion of the chest was chiefly on the left side. There was slight impairment of resonance on percussion over the left front and more definite impairment over the right front. Over the left upper lobe there was, on auscultation, a slight prolongation of expiration, but the sounds were almost normal. A few coarse rales were heard above and below the clavicle. The breath sounds were rather loud over the left lower front and axilla, but no adventitious sounds were present. Over the right upper lobe the breath sounds were very loud, with expiration prolonged, and on inspiration many coarse, dry rales. These were more numerous over the left upper lobe but still more numerous over the lower right lobe and in the right axilla, where they were heard throughout inspiration. Fremitus could be felt over both backs, but especially over the right. Over the right lower lobe the sounds were loud, but expiration bore a normal relation to inspiration. Down the whole right back the breath sounds were loud, with both inspiration and expiration vesicular and of normal relative duration. In the lower right lobe especially one heard a multitude of coarse, dry rales. The *x*-ray plates, taken by and interpreted by Dr. Albert M. Cole, show dense infiltration in the middle and upper portion of the right lower lobes, which extends well out to the periphery. The linear markings here are thickened. The greatest density is found along the hilum. The right apex is slightly more obscured than the left, but no trunks can be traced up into the right apex from the hilum. The entire left lung shows no roentgenological abnormality. There are practically no calcified glands in the hila and no shadows suggesting healed tuberculosis. This marked involvement of the right chest shows no roentgen characteristics of tuberculous disease.

The pulse was 64 to the minute, and regular in force and rhythm. The systolic blood-pressure was 126 and the diastolic 74 mm.

Heart. The point of maximum impulse was felt just at the nipple line. The deep cardiac dulness extended 11 cm. to the left in the fourth, which is 1 cm. lateral to the left mammary line, and 3 cm. to the right in the third interspace. The sounds were perfectly clear with the accent on the aortic second. There is now no edema of the ankles. Red blood count 5,720,000, and the white blood count 14,000 per cu. mm. Hemoglobin, 100 per cent. Differential count: small mononuclears, 17 per cent; large mononuclears, 8 per cent; polymorphonuclear finely granular cells, 67.2 per cent; eosinophiles, 2.2 per cent; basophiles, 1.4 per cent; and unclassified cells, 4.2 per cent.

Urine. The morning specimen was clear, acid, of straw color, and its specific gravity, 1022. It contained no albumin and no sugar. The sediment contained only a few epithelial cells.

The sputum is negative for Bacillus tuberculosis, but still contains the penicillium mould.

Wassermann test is negative; complement deviation for tuberculosis 50 per cent. For further laboratory work see page 12.

To review this case briefly, this patient's illness began seven years before his first admission, with a cough which sometimes was dry but which often raised a mass of mucus which occasionally contained "slugs." His smothering spells were clearly of intratracheal origin, so severe that he "felt he would choke to death," and required morphin for their relief. For several years he had been obliged to sleep in a chair. Although he was unable to work because of his respiratory distress, yet he had lost no weight, no strength, had not become anemic and had had but very little fever. He had never seen blood in his sputum and none of the numerous examinations had revealed Bacillus tuberculosis in it.

This man's dyspnea was quite unexpected considering his appearance of good health. The shape of his chest suggested both emphysema and fibrosis of the lung. That is, while rather barrel-shaped, yet it was flattened in front and expanded on deep inspiration but 13 mm. The apices of the lungs were relatively clear; his trouble seemed greatest near the hilum of the right lung. The percussion note generally was slightly impaired. The signs suggested a general bronchitis and fibrosis of the lung. These physical signs were out of all proportion to the fewer systemic symptoms. One must, of course, think of pulmonary lues, but we do not consider this likely. On the basis of this history and physical examination we at once remarked to the group of senior medical students with us that this probably was a case of mould infection and sent his sputum to the clinical laboratory for the benefit of the third-year class in clinical diagnosis and the patient to the *x*-ray department. The students had no difficulty in finding the mycelial threads and spores of a mould. Our roentgenologist, who, so far as we know, knew nothing of the patient, remarked later to the intern that the plates suggested a mould infection. The sputum was obtained several times by the most careful technic and cultures made, in all of which a penicillium mould grew. The patient was then given increasing doses of potassium iodide. During the subsequent six years he has

returned to the clinic for observation and also that we might demonstrate him to the class in physical diagnosis. He has improved considerably during these seven years, has been able to support himself by work practically all of the time. He said that from the first he had been able to get along without morphin. He is by no means well. He still has some dyspnea on exertion and some cough.

The second patient is a woman who has been for years treated for tuberculosis and whose sputum contains this same mould.

No. 10,396. Mrs. S., white, aged thirty-two years, admitted to the Robert W. Long Hospital, October 6, 1920, and discharged October 22, 1920.

Complaint. "Tired most of the time; morning cough with considerable sputum; frequent chilly feelings."

Family History. Father died, fifty years of age, of angina pectoris. Her mother, who is living, has always suffered from asthma. Has one brother, who is living and well, but no sisters.

Past History. Patient never had rugged health. She was ill five weeks with typhoid fever when twelve years of age, with a light case of whooping-cough at fourteen years of age and for one month with scarlet fever when twenty-one years old. She has had frequent attacks of tonsillitis since childhood. Her first pregnancy, ten years ago, was complicated by pernicious vomiting of such severity that labor was prematurely induced. This child now is in good health. That confinement was followed by a severe attack of fibrinous pleurisy over the lower left chest. The pleurisy pain was very severe, but was accompanied by very little fever and no effusion. The following year was spent quietly in California, taking the tuberculin treatment. She gained very slowly. Since then she has been considered a case of tuberculosis, although no tubercle bacilli have been found in her sputum, which also has never contained blood. She said that she never accepted this diagnosis, since "she felt too well."

In 1915 her second baby was born. During this pregnancy also she suffered from pernicious vomiting. This baby died of pyloric stenosis. About ten days after this confinement there developed a high temperature and then thrombosis of both legs. She then was confined to her bed for six weeks. Her limbs since then often swell and ache. Following this confinement she became a "nervous wreck," melancholic and with a morning cough and sputum.

We first saw her in 1917 after an attack of tonsillitis. She complained of a nervous aching all over the body and a severe pain in the back, legs and arms. She was depressed, especially during the noon hours. She complained that she often caught cold and had an attack of "grippe" and tonsillitis each year. She also suffered with gas in the lower abdomen, constipation, bloating, mucous colitis, leucorrhea and nervous feelings. Her

skin was too dry and cold. Her weight was constant. She had a morning cough and complained of palpitation of the heart.

She then (March 13, 1917) was fairly well nourished, the pulse 68 to the minute, the systolic blood-pressure 98 and the diastolic 86 mm. Hg. The lungs were fairly normal, save that expiration was prolonged at the left apex. Deep cardiac dulness extended 9 cm. to the left in the fourth interspace and 3 cm. to the right in the third. The heart was negative. The abdomen was negative. There was slight edema of the ankles. The tonsils were removed during the next month. On April 30, 1917, her doctor reported that the patient had done splendidly since this operation.

On October 29, 1917, she sought relief from constipation. At that time she was a little pale; her weight was constant; her appetite good; she had no indigestion; she coughed a little in the morning but raised no sputum. She always felt muscularly tired in the legs and back from the waist down, and was very depressed, especially in the afternoon when fatigued. The pulse was 64 to the minute, regular in force and rhythm; the systolic blood-pressure was 114 and the diastolic 76 mm. Hg. The lungs now were clear at the apices; the abdomen was normal; the veins of the legs were slightly distended; there was no edema. Pelvis normal.

X-ray examination showed that her constipation was due to a general colonic stasis, that it was not of the rectal type and not due to pressure of the retroflexed uterus. She was given lutein tablets. On December 7, 1917, she reported that she was much better.

Present Illness. The group of symptoms which interests us now began January, 1920, as easy fatigability, palpitation of the heart, tachycardia (120 to the minute), shortness of breath and morning cough with considerable sputum. She raised no more sputum during the rest of the day. She had lost weight (ten pounds). Her skin was dry; she chilled easily; the temperature in the morning ranged between 96° and 97° and in the afternoon between 98° and 99.3.° Systolic blood-pressure April 17, 1920, was 113 and the diastolic 80 mm. Hg.

Lungs. The movement on respiration of both upper lobes, but especially the right, was limited and their resonance on percussion impaired. The breath sounds over most of the upper chest, but especially the right, had a tubular modification. Over the right upper and middle lobes and over the left upper lobe one heard an unusual number of rales of several varieties, but especially the sonorous and medium moist rales. The heart was negative.

April 17, 1920, the sputum was very thick and tenacious, ropy, mucopurulent and of a brownish color. It contained many pus cells, a few red cells, a few mycelial threads, no yeast cells, no elastic tissue and no tubercle bacilli. This sputum had not been obtained under aseptic conditions, therefore we disregarded the mould threads.

On October 5, 1920, after five months' treatment in Colorado, she came again for examination. Most of this time she had been in bed, but had had no fever, and the thirty or more examinations of the sputum for Bacillus tuberculosis all were negative.

A letter from Dr. A. M. Forster, dated September 30, 1920, read, in part, as follows: "My examination after admission showed the following: *Right side*, dulness to fourth rib and to eighth vertebral spine, with bronchial breathing to second rib and to third vertebral spine; prolonged high-pitched expiration to fourth rib and sixth vertebral spine; medium coarse and fine rales to fifth rib and over the back; *left side*, dulness to third rib and sixth vertebral spine, with prolonged high-pitched expiration; medium coarse and fine rales to fifth rib and eighth vertebral spine. Subsequent examinations have shown definite clearing up of the rales, but, as you will note, they are still very numerous and cover a wide area. Her temperature and pulse-rate have been practically normal since she has been under our observation. Her *x*-ray plate does not show the characteristic changes one would expect to find from so extensive a tuberculosis as her physical signs would indicate. This together with her history of repeated examination, *x*-ray and physical, by competent men, and also her lack of acute symptoms and the fact that she has had negative sputum, have made us hesitate to feel that her condition is due to tuberculosis alone. We have told her that she has a "chronic lung infection."

Her pulse on this date (October 5, 1920) was 88 to the minute, regular in force and rhythm; systolic blood-pressure 105, and diastolic 68 mm. Hg. The pulmonary condition was that described in Dr. Forster's letter. The rales over both upper lobes were very numerous. Menstruation then was very profuse. She had a morning cough and some sputum, also a bloody nasal discharge, but *x*-ray examination of the nasal sinuses was negative. The character of these lung signs now suggested mould infection so strongly that we sent her to the Robert W. Long Hospital for careful study. The *x*-ray plates taken by Dr. Cole showed marked hilum involvement on both sides, but no roentgen characteristics of tuberculous disease. Both apices were clear. There were practically no calcified glands or scars anywhere in the chest. The greatest involvement was at the right border of the heart, where there was a marked density with thickened trunks running out toward the periphery. This picture suggested a pulmonary mould infection.

During the seventeen days she was in the hospital her temperature reached 99° but five times, the highest record being 99.4°. The pulse varied from 64 to 96 to the minute with the mean rate between 76 and 80. The systolic blood-pressure was 96 and the diastolic 70 mm. Hg. On admission the patient weighed 123 and when discharged 124.5 pounds. During this stay in the hospital the Goetsch test was used to determine whether or not some of her symptoms were due to hyperthyroidism, but no reaction followed the subcutaneous injection of 8 minims of adrenalin.

Urine. October 6, 1920. The morning specimen was acid, clear, had a specific gravity of 1015, contained no sugar and but a faint trace of albumin. The sediment contained a few epithelial cells but no casts.

Red blood count, 4,860,000; hemoglobin, 80 per cent; leukocytes, 8,800, of which 28.7 per cent were small mononuclears, 4.3 large mononuclears, 66 per cent polymorphonuclear finely granulars and 1 per cent eosinophils.

The sputum was examined October 7, 8 and 9 for tubercle bacilli, elastic tissue and red blood cells, but none were found. Repeated cultures of the sputum were made, using the most careful technic. (The mouth and teeth were well cleansed the night before and the morning of the test. The patient expectorated into a sterile beaker. The sputum was then thoroughly washed and cultures made.) A penicillium mould grew each time. The stools were negative.

Potassium iodide in increasing doses was prescribed.

(For other laboratory reports see page 12).

October 30, 1920. The patient now has morning headaches and backache at waist level. Her temperature is below normal every afternoon, even 96.6° at 4.00 P.M. She still expectorates each morning, but the sputum has not been increased by the potassium iodide therapy.

November 21, 1920. The patient now has a fever each afternoon, especially if she lies down, but not if she is up. She is "tired to death," and has lost 1½ pounds in three weeks. Systolic blood-pressure is 96 and diastolic 76 mm. Hg. She coughs a little and has some morning sputum. This morning sputum now rises in one mouthful, which almost chokes her. She raises very little during the rest of the day. We advised that she return to Colorado Springs.

September, 1921. The patient returned from Colorado Springs much better.

Reviewing this case we would call attention to the fact that for years she has been treated for tuberculosis, notwithstanding the negative reports from clinical and roentgenological laboratories. Drs. Webb and Forster, at Colorado Springs, recognized the atypical features of her case. We considered her a case of healed tuberculosis with considerable pulmonary fibrosis, until one day, two years ago, when the pulmonary signs became even startling and suggested the rapid extension of a tuberculosis, although she said that she felt not a bit worse, only easily fatigued, and she coughed a little oftener than before. The fact that her afternoon temperature touched 99° a little oftener than usual was the reason she came then for examination. On auscultation—rales were heard throughout

the entire chest, but especially on the left side, a multitude of rales, especially the medium and fine moist and some of the dry varieties. The apices were not especially involved. We sent her back at once to Colorado Springs, to Cragmont, where she stayed five months. During this time at least thirty routine examinations of the sputum for tubercle bacilli were made, all of which were negative. She had no fever, but the pulmonary physical signs remained unchanged. Becoming restless, she returned to Indiana with a diagnosis of fibroid phthisis with secondary infection. The fact that we now found the physical signs practically the same as when she had left, five months before, made us now suspicious of mould infection. The roentgenologists were then very willing, from a study of the plates, to make a diagnosis of mould infection of the lung, while the laboratory men grew from the sputum a penicillium mould the growth of which resembled Penicillium crustaceum. Its spores measured from 2 to 3.1 microns in diameter. We prescribed iodides for continuous medication and later sent her back to Colorado Springs. While she is by no means well, yet she is improving.

These two patients have certain points in common. Their infections are of years' duration and have been considered tuberculous. Their infections have had very little influence on their general health; they have not lost weight or strength; they have felt well; they have been almost afebrile. The pulmonary signs on inspection, palpation and percussion have been those of fibroid phthisis, but auscultation has suggested a serious, rapidly extending, general tuberculosis. It was positively uncanny to listen to the multitudes of rales in the chest of patients so little sick. The question now arises, Have these patients pulmonary tuberculosis? or, granting that all may have a little, are the symptoms and signs described above due to pulmonary tuberculosis? Undoubtedly we have in past years been far too willing to pigeon-hole as pulmonary tuberculosis all those cases whose symptoms strongly suggested this and have not always waited until the proof was positive. We have had no hesitation in sentencing to the far west, for a stay of one or two years, any young person whose symptoms and signs even suggested tuberculosis. Recently, however, a reaction fortunately has set in and we are beginning to realize that the diagnosis of pulmonary

tuberculosis is a very difficult matter. We now study our cases long and seriously before committing ourselves. It is interesting that Castellani reported that of all the Italian soldiers referred to him in the past war for pulmonary tuberculosis at least 3 per cent. were cases with mycosis or sporochetosis of the bronchi. Nathan evidently had the same opinion, for he says that many cases of so-called tuberculosis are mould infections and that the mistake is never discovered, since these patients are referred to hospitals for the tuberculous, where they soon contract that disease, thus confirming that diagnosis.

In these two cases all of the many previous routine sputum examinations, made by several others and by ourselves, had been negative for Bacillus tuberculosis, elastic tissue and blood. We now repeated these examinations, using the most careful technic, but found none. Cultures of the sputum for Bacillus tuberculosis were made. These proved negative. Guinea-pigs were inoculated with the sputum. All of these attempts to demonstrate Bacillus tuberculosis in the sputum failed.

Biological tests were next used—not the tuberculin reaction, since, positive or negative, it proves so little, but rather the complement-fixation test for tuberculosis. In this we were assisted by Prof. Virgil H. Moon, who has been much interested in that reaction. The man reacted weakly positive, but not more so than many normal persons, while the woman's reaction was quite negative. The roentgenograms not only do suggest mould infection but do not suggest tuberculosis. That is, there are no calcified glands at the hila of the lungs; the apices are relatively less involved than the rest of the lungs; the lesion was a diffuse infiltration, densest at the hilum and radiating to the periphery. One sees no typical ramifying and anastomosing thickened bronchi; no nodes along the bronchi, etc. And, lastly, if these patients are tuberculous, they certainly have thrived on a continuous potassium iodide treatment.

The next question is—are these cases of mould infection, or is the mould merely a contamination or an accidental member of a mixed infection? In reply it may be said that the clinical courses of these cases are not those of tuberculosis nor, again, of pyogenic pulmonary

infections; they resemble those of other primary yeast and mould infections. The systemic symptoms have been few; the fever slight; there is little loss of weight and strength; and they have improved on iodide treatment. Again, the physical signs were not those of chronic fibroid phthisis. The *x*-ray plates showed clearly the lesions of a pneumonomycosis; the sputum obtained under the best of technic never contained Bacillus tuberculosis nor blood nor elastic tissue, and always did contain the mould. Controls were made in the rooms where the work was done and they all were negative for moulds. The physical examinations and the *x*-ray plates failed to show any pulmonary cavities or dilated bronchi which could harbor a saprophytic organism.

Had the mould been an aspergillus the cases would not be challenged, for that group of mould is accepted as pathogenic; but is the penicillium pathogenic? We have found but one case in literature, that of Canio, who reported an acute case from whose sputum he grew the penicillium. Dr. Moon and his assistant, Mr. Frank Forry, took up the research for us. Canio reports that the injection of the sputum of his patient into the peritoneal cavity of laboratory animals caused lesions. The injection of our patient's sputum into the peritoneal cavity of a guinea-pig produced nodular lesions in the mesentery and omentum which we believe were due to this mould, but when they injected the pure cultures of the mould into the peritoneal cavity and into the lung of rabbits they got negative results. This is what our knowledge of mould infections in general would lead us to expect. Mr. Forry then attempted by injecting increasing doses of a suspension of the spores to immunize rabbits to this mould, but was unable later to demonstrate in their blood the presence of any complement-binding body. Whatever pathogenicity this mould may have it evidently owes to some relationship to other organisms. Dr. Elmer Funkhauser, of Dr. Moon's department, studied the other organisms of this washed sputum and found staphylococci, non-hemolyzing streptococci and Micrococcus tetragenus.

We believe, therefore, that in the mixed pulmonary infections of these two patients a penicillium mould plays some and that

Bacillus tuberculosis plays no part. Of course we are on the lookout for other cases, since it may be that mould and yeast infections are more definitely regional in their distribution than is generally believed. Certainly an unusual number of cases of blastomycosis are found in Chicago; of oïdiomycosis in California; of Oïdium brasiliense in Brazil, etc. It may be that in Indiana there are more penicillium cases than elsewhere.

DISCUSSION

Dr. L. G. Rowntree, Rochester, Minn.: Does sodium iodide have any effect in these cases? There is a tremendous difference in the toxicity of potassium iodide and sodium iodide. Some syphilographers are using relatively tremendous intravenous injections of sodium iodide. For instance, Dr. Stokes, at the Mayo Clinic, routinely uses as much as 100 to 200 cc of 10 per cent sodium iodide solution intravenously daily. If there is any response to sodium iodide the possibility of using this larger dosage might be of value.

Dr. J. B. Herrick, Chicago, Ill.: I should like to ask if Dr. Emerson noted any particular features in the *x*-ray plates; if so, what were they?

Dr. George Blumer, New Haven, Conn.: Did they use tuberculin in those cases?

Dr. T. R. Boggs, Baltimore: I should like to draw attention to a very important reason for non-discovery of mould infections of the lung. We have got away from studying the sputa in gross, and I have convinced myself that it is easy to overlook massive infections in sputum when one only uses the ordinary staining methods with anilin dyes. The mycelium is so distorted by the heat and fixation that it is not recognizable. The spores stain imperfectly and are supposed to be large cocci. I think we must study the sputum first in gross and then by microscopic methods or we shall continue to overlook these cases, which must be vastly more frequent than anyone realizes. The use of black and white surface on glass, of the loop and study of suspected particles on acid carbohydrate media that will restrain growth of other organisms and permit development of the fungi are essential points. In regard to Dr. Emerson's point—recognition of fungus infections in the tissues—it is not easy by any section or fixation

method. One may look through any number of sections of a gland stained with eosin or hematoxylin and see no evidence of mycelium, although it may be present. We must pay special attention to the methods of demonstrating these infections, and must look to the bacteriologist for some serious effort to standardize the present chaotic descriptions of these organisms. If one goes through the literature it is difficult to tell whether writers are describing the same organism or not. It is the technical bacteriologists who know about moulds. Medical men do not know how to describe them technically. Only after exhaustive cultural and morphologic study of a given organism is it possible to state accurately the exact classification.

Dr. S. Solis-Cohen, Philadelphia: Carrying out the statements of the last speaker in regard to laboratory methods in some laboratories: When non-tuberculous sputum is sent in one commonly gets the report, "No t. b.," and nothing else. If the laboratory is specifically requested to note what is present as well as what is absent the report may sometimes come, "Unknown organism." Something more than the ability to prepare a culture medium or to stain a slide is necessary for the technician. The growing tendency to turn this work over to persons without medical or at least pathologic training is a serious error.

Dr. W. S. Thayer, Baltimore, Md.: I had two instances of sporotrichosis under my observation, one of which I had intended to present last year.

A woman came to the hospital in the fall of 1916 presenting a picture of tracheal or bilateral bronchial obstruction and dyspnea. She had been sent by her physician to Dr. Minor, of Asheville, who had been convinced that the condition was not tuberculous and suspected some mould infection. She showed with us evidences of induration at the roots of both lungs. There was nothing definite in the sputa. The Wassermann reaction was + + + +. She returned to her physician and was put under vigorous antiluetic treatment with arsephenamin. She did not have iodide of potassium. On returning from the war I found the specimens from the necropsy in the laboratory. The lungs showed at both roots extensive wedge-shaped, slate-colored sclerosis; in these sclerotic masses the bronchi were clasped as in a vise. In the tissue itself we found sporotricha. These were cultivated, but the group to which they belonged was not positively identified. The sputa while in the hospital were unfortunately scanty and I fear that the examination of the fresh specimen was neglected. It is not impossible that neglect to examine the fresh sputa, as important as the examination indeed of the fresh blood, may account for our failure to recognize more cases.

In another instance a colored man showed an acute respiratory infection with some subcutaneous nodule-like subcutaneous gummata. It was not

luetic. There was no definite evidence of tuberculosis. Sporotrichosis was suspected and the patient after thorough treatment with iodide of potassium has improved somewhat, but we are not encouraged as to the outlook. There is no clear evidence of pulmonary disease. From one of the subcutaneous gumma a sporotrichium was cultivated on one occasion by Dr. Guthrie. Microscopically the organism was not found and Dr. Guthrie is not convinced that the growth may not have been a contamination.

The case now under observation is, however, of interest.

Dr. Charles P. Emerson (closing): In answer to Dr. Rowntree's question, I would say that we prescribed potassium iodide. We have the *x* ray plates here for you to study. The picture is quite different from that of tuberculosis. In that disease one more often finds a lesion of the upper lobe, longer bronchial markings with anastomoses and evidence of calcified glands near the hilum. The *x*-ray men, without knowing about the case and receiving no suggestion, thought it mould infection.

In answer to Dr. Blumer's question, we would say that we did not use tuberculin. The evidence of tuberculosis based on the subcutaneous or conjunctival test is in adults so difficult that we have been elaborating our technic of the complement-fixation test for tuberculosis, which we believe more valuable. If we get a negative complement-fixation test for tuberculosis we had better negative evidence than had we used tuberculin. If we got a positive tuberculin test with either it might leave us in doubt.

We have found aspergillus cases, but these are penicillium cases, and we could find only one other case like these in literature.

A PHYSICAL BASIS FOR EPIDEMIOLOGY[1]

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(ABSTRACT)

In the autumn of 1918 there swept through one of the mouse-breeding rooms of the Rockefeller Institute a destructive epidemic of mouse typhoid—an infection of mice with a bacillus of the enteritidis group of organisms, to which the name of Bacillus typhi murium has been given. The history of the epidemic is instructive: The original mice of the population, numbering about 3000 at the time of the epidemic, came from a breeder in Massachusetts and had been purchased some time before and moved *en masse* to the Rockefeller Institute. In the meantime many new mice had been born of this stock and many of the original mice had died or been employed for experiment, so that only a small residue of the original population remained. In other words the epidemic of mouse typhoid arose among chiefly a new stock of mice, the offspring of an old stock believed on good grounds to have passed through previous outbreaks of the disease.

There is still another reason for supposing that the epidemic arose from within and was not imported from without this stock. Besides the breeding-room for the population accruing from the purchased mice, a second breeding-room not far away is maintained for mice bred at the Institute from perfectly healthy stock. This second

[1] From the Laboratories of the Rockefeller Institute for Medical Research. A complete report will appear in the Journal of Experimental Medicine.

breeding station has been developed from small beginnings and now has an average mouse population of 3000. The personnel in charge of each breeding room is distinct and does not mingle. During the period of many months in which the epidemic continued, rose and fell in the one room no death from mouse typhoid occurred in the other.

Our attention has long been directed to the phenomena of the epidemic prevalence of disease, and since the early years of the founding of the Rockefeller Institute opportunity was presented to observe in succession epidemics of meningitis, poliomyelitis, influenza and latterly of encephalitis. The waves of these diseases, which have swept over the world since 1904, have aroused a profound interest in the facts of epidemiology.

We seized, therefore, upon this outbreak of mouse typhoid in order to study experimentally an epidemic disease among small laboratory animals which could be assembled and observed in fairly large numbers. The epidemic referred to has supplied the cultures of the Bacillus typhi murium for the experiment and certain data of a statistical nature with which to compare our experimental results. The healthy stock of mice also referred to provided unexceptionable material with which to attempt the production of an intentional epidemic.

The conditions of the experiment were simple: A kind of mouse village was set up and an isolated room away from all other animals was selected in which shelves were erected and the cages placed. The shelves and cages may be taken to represent streets and houses in an ordinary village. Each cage contained five healthy mice. The original population, later increased from time to time by the introduction of fresh increments, was 100. The epidemic was started by feeding with a virulent culture of Bacillus typhi murium ten mice placed in two cages midway of the other cages. The incidental contact between the culture-fed mice and the others was secured by the attendant who fed the animals and periodically cleaned the cages. A spot map was kept in order to follow the events.

The preliminary feeding experiments led to the death from mouse typhoid of eight of the ten culture-fed mice and of seven of the

contact mice distributed in as many cages. No epidemic in the real sense ensued. The fatal outbreak of mouse typhoid which arose partook of the nature of sporadic instances of the disease.

When the conditions as regards death from the infection had become stationary (at about the end of 30 days) a fresh addition of 200 mice contained in forty cages of five each was brought into the village. The effect of this addition was striking: After a lapse of about five days the new mice began to die of mouse typhoid, and five to ten days later the old mice (original population) also began to succumb. The total losses in this outbreak were 70 per cent and the number of cages involved, all, or 100 per cent. The epidemic endured about four weeks and then subsided, although an occasional death still occurred at intervals.

When a second equilibrium had been established another addition of normal or healthy stock was introduced into each cage of the village to replace those which had died during the preceding period. Thus after recruiting, the total number of mice in the village was 300. The succession of events was about the same as that just described. However, the peak of the epidemic was less high, the total percentage of deaths being 58 per cent, although 100 per cent of the cages was again attacked. After an interval another equilibrium was reached, when still another addition of normal mice brought about a recrudescence, passing, as in the other examples, from the healthy new population to the exposed old population, with, however, a total percentage of deaths below the others.

These fluctuations or movements of the epidemic, consisting of a series of waves, have been repeated now about ten times. The general character or type of the wave or curve is always the same, although the height or peak as determined by the percentage of mortality varies. In every instance the death-rate among the old mice (*i. e.*, those exposed to preceding epidemic periods) had finally equalled the rate among the new mice introduced.

The experiments are followed in two main ways: According to the mortality, controlled, of course, by bacteriologic examination of the animals succumbing and according to the living carriers of the Bacillus typhi murium produced. The feces and the urine of living mice are tested for the bacillus, and in general it may be

stated that the percentage of carriers stands in inverse ratio to the mortality. In other words, high death-rate means a low carrier rate, and *vice versa*. An obvious, although by no means safe, deduction from this observation would be that the carriers have all recovered from non-lethal infection, and hence are rendered resistant by an acquired immunity and that the production of carriers is Nature's way of limiting the epidemic and of bringing about the state of equilibrium described.

The main results of the study presented may be viewed as the mere beginnings of an undertaking to follow artificially reproduced epidemics among animals which not only should simulate the epidemics naturally or spontaneously, as we say, arising among men and animals, but which, being controllable, may be made to yield up some of the underlying conditions affecting the movements among those naturally occurring epidemic outbreaks which we represent graphically in the form of curves but of which in relation to cause and effect we still are so largely ignorant.

The impulse to use epidemic diseases among laboratory animals to help solve the problem of epidemics among man has been felt by other experimenters. In particular, Topley,[1] in London, has independently made use also of the mouse and a bacillus of the enteritidis group with which to conduct experiments, and the reports he has already published, in spite of quite fundamental differences of procedure, are readily comparable with those we have obtained.

As a matter of practical importance perhaps the epidemic diseases affecting the respiratory tract may be more important than those in which the portal of entry into the body of the infectious microbe is by way of the digestive tube. The latter class has indeed been subject to far greater control by sanitary and other measures than the former. But in endeavoring to work out the underlying principles ultimately to be assembled into the "laws" of epidemics, information from many sources will be required. It is part of our object, however, to report later on experimental epidemiological studies now in progress with a disease of laboratory animals in which the infecting bacillus thrives on the respiratory mucous

[1] Journal of Hygiene, 1921, xix, 349.

membrane. It is obvious that the two chief factors involved in epidemiological investigations, namely, the host and the microbe, are both highly intricate. Any comprehensive study will include both, and to the extent to which it is enlightening will account for the effects of the fluctuation of one on the response of the other, the two conditions having been correlated, providing the data on which our ultimate understanding of epidemics will come to rest.

DISCUSSION

Dr. Simon Flexner, New York: I should like to explain why we undertook this work. We have been greatly impressed with waves of epidemics in this country. We have had poliomyelitis and we have had influenza, and lately epidemic encephalitis. The study of these waves has been restricted to statistical data from human beings. At the Rockefeller Institute we had a very violent epidemic of typhoid in our cancer-breeding mouse stock, which is a separate stock. We thought we would try to get experimental light on the epidemic and on the curves of epidemics as ordinarily produced. We began to study our results and we are extending this study to one of respiratory infection in rabbits, commonly called "snuffles." We shall report on this later. These two studies cover epidemics of gastro-intestinal infection and respiratory infection. The facts which have come out are very interesting, especially as regards the introduction of normal mice among the population which has successfully passed through the epidemic. Dr. Topley, of London, has undertaken a study which is in general agreement with what we have considered, and though there are some differences, and he has some points which we have not, while we have some that he has not, there is a general coincidence. These studies differ from those carried on among human beings in that they deal with mortality entirely, while human studies deal only with morbidity. We do not know whether the mouse has the infection unless it dies, so that we cannot draw exact parallels between this and human infection, but they may be compared with mortality statistics. The striking thing is the wave incidence, even when measured entirely by mortality. We thought it wise to present these studies at this juncture, because we expect to go on for many years and study different phases, and hope to acquire data referring to the sources and causes of epidemic waves of certain diseases so numerous in the literature.

REMARKS UPON THE SYMPTOMATIC TREATMENT OF PNEUMONIA*

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The hope for a true curative treatment for the various types of pneumonia undoubtedly lies in the field of specific therapy. There are, however, but few successful forms of specific therapy in all medicine, and for the most part the successful practice of the art of medicine has lain in the past in the direct treatment of signs and symptoms, and in spite of the more brilliant results obtained with specific remedies, it cannot be denied that symptomatic treatment has met with no small measure of success.

At the present moment it is our object, while waiting for the perfection of the specific cure, to consider the matter of the symptomatic treatment of pneumonia, with special reference to the attempt to find the true indications for treatment in terms of morbid physiology. Symptomatic treatment may be either empirical or rational—the latter, of course, is the method of choice. Rational symptomatic treatment is only possible when not merely the anatomic but also the functional derangement in a given patient can be clearly visualized by means of knowledge gained from symptoms and physical signs and from properly understood laboratory and functional tests, and when, from such a visualization, therapeutic measures of known effect are employed to meet truly existing indications.

To apply such an ideal to the treatment of pneumonia it becomes

* From the Medical Service of the Massachusetts General Hospital. Aided in part by a grant from the Proctor Fund, Harvard Medical School.

necessary first to analyze the functional difficulties with which the pneumonia patient is confronted.

The outstanding difficulty is the respiratory battle. Even in the absence of specific remedies it seems likely that a considerable number of persons who now succumb to pneumonia could be saved were this respiratory burden taken from them. The ability to overcome toxemia and to repel bacterial invasion is undoubtedly reduced by exhaustion. The majority of pneumonia patients recover as it is. It would therefore seem reasonable to suppose that were the respiratory burden removed and exhaustion from respiratory effort prevented or decreased the mortality could be reduced to a very low rate indeed. Given half a chance the pneumonia patient will of himself survive his toxemia and master his infection. It will be our effort to show that there are methods at hand which it may be hoped will give him the half-chance by removing or diminishing his respiratory burden.

Difficult breathing, or dyspnea of a more or less typical type, is the prime subjective and objective respiratory manifestation of pneumonia. Let us consider, therefore, the manner of its causation. It has long been known to be at least not solely explainable on the basis of morbid anatomy. The extent of the pulmonary consolidation bears no constant relationship to the intensity of the dyspnea, and, furthermore, dyspnea may cease when the crisis is past with no alteration in the anatomic process in the lungs. We must seek then a physiologic as well as an anatomic explanation.

Dyspnea occurs in normal persons under certain circumstances, after violent exertion, for example, or when the oxygen in the inspired air is greatly reduced, as at high altitudes. It must be borne in mind that the term dyspnea should be restricted to difficult or distressful breathing. It is essentially a subjective phenomenon, though usually in addition it is possible to tell objectively when the breathing has become distressful. Increased breathing to which the term hyperpnea is properly applied is not necessarily distressful. One may have hyperpnea with no dyspnea. Hyperpnea is a condition in which the total volume of air entering the lungs per unit of time is increased over normal when, in other words, the ventilation of the lungs is increased. Increased ventilation of the

lungs may be accomplished by an increase in rate or in depth of respiration, or more commonly by an increase in both. An increase in rate of breathing only is not necessarily an indication of hyperpnea. Types of rapid shallow breathing occur in which there is no increase in the ventilation of the lungs, at least not in alveolar ventilation. To such the term tachypnea may well be applied. Tachypnea would be the respiratory homologue of tachycardia, and may, as in some tachycardias, be due to purely reflex or nervous causes. The work of Peabody and his collaborators,[1] and also that of Meakins,[2] has thrown considerable light on the nature of these several respiratory types.

With these definitions in mind we are in a position to discuss the factors which may cause dyspnea. In the first place in normal individuals we have a great variation in the amount of exertion necessary to produce dyspnea. The trained athlete may perform, with no subjective breathlessness, an amount of work which would produce extreme dyspnea in an untrained but otherwise normal person, and an individual with the so-called effort syndrome may have dyspnea on very trivial exertion. What are the reasons for these differences and what, in fact, is the essential cause of dyspnea?

We think that, in general, it can be laid down as axiomatic that in any individual, normal or diseased, the symptom dyspnea will arise whenever the pulmonary ventilation called for by his life processes at the moment exceeds the quantity of air that his pulmonary bellows is mechanically capable of delivering with ease. If this principle is sound the onset of dyspnea in persons, either normal or abnormal, will depend upon the ratio between the ventilation called for by bodily needs and the ventilation that their bellows can deliver. It is really a case of demand and supply. In the case of the athlete doing increasing amounts of work the demand that is to say—the call for pulmonary ventilation—is essentially the same as in the untrained subject, but the supply—that is to say the ventilating power of his pulmonary bellows—is greater. The result is that the athlete gets dyspneic less readily. The reverse holds in the man with effort syndrome. His supply is lower than normal; he gets dyspneic sooner. The difference in the ventilating power of the lungs in different individuals is largely dependent upon their

vital capacity. The tidal air can obviously never exceed the vital capacity; as a matter of fact it never reaches it. To make each individual respiration maximal would be a fatiguing and uneconomic way of producing ventilation. A high rate could not be maintained under these circumstances, as it takes time and effort to make a maximal respiration. The ventilating capacity will also be affected by certain other factors. The demand for ventilation on the part of the body is for a certain alveolar ventilation, or for a certain amount of air actually to enter the lung alveoli. The supply of ventilation—or the amount the bellows can deliver —must be thought of in terms of total ventilation. If the dead air space is increased a larger total will be necessary to accomplish a given alveolar ventilation. Also, rapid shallow types of breathing will require a greater total for a given alveolar ventilation than will slow deep types. The rapid shallow type is, in other words, an uneconomic type. These types have been discussed by Edsall[3] and their significance in determining the relationship between ventilation demand and supply must be borne in mind.

During progressively increasing muscular work there is in normal subjects, as has been shown by Boothby[4] and by Means and Newburgh,[5] a progressive and essentially parallel increase in pulmonary ventilation, gas exchange and blood flow. In the course of this progression dyspnea arises when ventilation reaches a point that throws a strain on the ventilating capacity of the lungs, and inability further to increase the work will be encountered when the subject reaches the greatest ventilation or circulation rate of which he is capable. The total ventilation of well-developed normal men may be increased to as much as eight or nine times the resting value. The upper limit, as shown by Peabody,[6] will be greater the greater the vital capacity. The increase in total ventilation is met, as has been said, by an increase in both rate and depth of breathing, but the increase in depth rarely exceeds a third of the vital capacity under any circumstances.[7] In producing large ventilation, therefore, the person with a small vital capacity will be at a disadvantage.[8]

Let us now apply these principles to pneumonia. We must again consider the demand for and the supply of pulmonary ventilation. In the normal person the demand is governed essentially

by the gas exchange, that is to say, by the metabolism. Thus, as we have said, with increasing muscular work the gas exchange and ventilation increase in essentially direct proportion. In this case it is the metabolism which is cause and the ventilation which is effect. The respiratory center wishes to maintain a constant alveolar CO_2 tension. To do this it can easily be seen ventilation must increase in like proportion to CO_2 output. If it did not, alveolar CO_2 tension would be altered.

In a disease such as pneumonia the metabolism will, as in the normal, be one of the factors determining the volume of the pulmonary ventilation; an increase in metabolism due to the disease will call for an increase in ventilation exactly as the elevated metabolism of muscular work did in the normal person. There are recognized in disease causes for increased metabolism which are quite apart from bodily activity. Fever is one of them. An increased level of the metabolism has been shown to exist by Du Bois and his collaborators in the fevers of typhoid,[9] malaria[10] and sometimes in tuberculosis.[11] The typhoid patient at the height of his disease may have an increase of 40 to 50 per cent in his metabolism resulting from his fever alone and not due to muscular activity. It is altogether probable that a similar increase occurs in the fever of pneumonia. The metabolism, then, of the pneumonia patient may be expected to be higher, even while he is at complete rest than it would be under the same conditions when he was well. He will, in other words, have a metabolic need for increased breathing or hyperpnea.

The metabolic, however, is not necessarily the only factor calling for hyperpnea. Another possible one is acidosis. If blood alkali is used up by combination with acid, in order to preserve blood reaction at its normal point, the carbon-dioxide tension of the blood must be reduced. This can only be accomplished by reducing the alveolar CO_2 tension through increased pulmonary ventilation. In general terms it may be said that to lower the alveolar CO_2 tension to half its original level the ventilation must be doubled.[12] The presence or absence of acidosis can best be determined by constructing the so-called carbon-dioxide diagram of the blood.[12] To do this one determines and plots the carbon-dioxide dissociation

curve. The blood is equilibrated with three atmospheres, each having a different and known tension of carbon dioxide but the same tension of oxygen. The carbon-dioxide content of the equilibrated blood is determined by blood gas analysis. The three points obtained are plotted and a curve drawn through them as in Fig. 1, the contents (expressed in volume per cent) being the ordinates and the tensions (expressed in millimeters of Hg) the abscissæ. Now the reaction of the blood, Henderson[13] has shown, depends chiefly on the ratio between the concentration of free carbonic acid and of bicarbonates, thus:

$$[H^+] = K\frac{[H_2CO_3]}{[BHCO_3]}$$

in which $[H^+]$ is the hydrogen-ion concentration (and hence the reaction) of the blood $[H_2CO_3]$ the concentration of carbonic acid and $[BHCO_3]$ that of bicarbonates in the blood and K a constant. The reaction of the blood at any point in our dissociation curve can be derived therefore from its ordinate and abscissa, for the ordinate, or carbon-dioxide content, is proportional to bicarbonate concentration while the abscissa, or tension, is equivalent to the concentration of free carbonic acid, for gases go into physical solution in direct proportion to their tensions.

This being true, in the diagram any diagonal line drawn through the point O will represent a certain hydrogen ion concentration, or blood reaction, for the ratio of ordinate to abscissa for any point in such a diagonal will be the same. By the formula of Haggard and Henderson, Peters, Barr and Rule[14] have constructed such diagonals for different blood reactions. They are shown in Fig. 1. The logarithmic expression of hydrogen ion concentration, the so-called P_h, is usually used.

Having plotted the dissociation curve of a given blood one can then obtain samples of blood directly from an artery or vein and determine their CO_2 content by analysis and then plot these on the previously plotted curve. These are called the arterial or A point and the venous or V point. From the position of these points the reaction of the blood can be read off on the diagonal.

We have to date constructed twenty CO_2 diagrams for the bloods of thirteen patients with pneumonia. Three of these were shown to this Association a year ago.[15] The rest have been determined since then. We also have to date a series of some eighteen diagrams of controls of various sorts. It was pointed out in the previous communication that acidosis in the sense of an abnormal amount of non-volatile acid in the blood is shown by the level of the dissociation curve. A low curve means that a portion of the blood alkali is used up. The presence of abnormal acid is usually cared for and compensated for by reduction of carbonic acid through hyperpnea. When this compensation is sufficient, even though the dissociation curve is low, the A and V points will fall on the diagonal of normal P_h (which is 7.35). When compensation has not been adequate there is a shift in blood reaction, and this will be shown by a position of the A point or V point to the right of the P_h 7.35 line, which is in the direction of acidity.*

In the previous paper[15] there was shown a zone within which it was expected normal curves should fall. The majority of our curves for pneumonia patients fall within this. A few fall slightly below it. For this reason we believe that often there is no non-volatile acidosis in pneumonia, or when present it is of slight grade.† When we come to the P_h of the blood in pneumonia we find that certain cases show a reaction less alkaline than normal, even though the dissociation curve is at a normal level. This, to our minds, can mean but one thing, a retention of carbon dioxide giving rise to what may be called a carbonic acidosis. In normal persons the alveolar air and arterial blood are essentially in tension equilibrium. This, as Peters has shown, is not necessarily true in disease.[16] In pneumonia or in heart disease it may often happen that all the blood does not get properly aërated as it passes through the lungs. This may be because of consolidation, edema or to exudate or foam in the air passages, as suggested by Hoover.[17] Under such circumstances the blood leaving the lungs will be a mixture of aërated and unaërated blood. Furthermore,

* P_h falls as a solution because less alkaline or more acid.

† These curves will be published in full and discussed in detail in the near future by Means, Barach and Woodwell.

the CO_2 tension of the blood will be higher than that of the alveolar air. If in a portion of the lungs a proper gas exchange cannot take place, in order to maintain blood CO_2 tension at a normal level the normal portion of the lungs must be overventilated. Impairment, then, in the respiratory function of any portion of the lungs, if it leads to a mixture of aërated and unaërated blood, will be a factor demanding hyperpnea. It may well exist in pneumonia.*

A third possible cause for hyperpnea, which has recently been suggested by Pearce,[18] and which may at times be present in pneumonia, is an insufficient circulation rate or blood flow. We pointed out before that as metabolism increases ventilation and blood flow must increase in like proportion. To a certain extent, however, as Pearce shows, an insufficient response on the part of the circulation can be offset by an increased response on the part of the ventilation—superventilation as he calls it. From this it would follow that in an individual whose circulation rate did not increase parallel with his metabolism, dyspnea would occur at an earlier point because of the superventilation which compensates for the insufficient blood flow. In a pneumonia patient with poor heart action and insufficient blood flow, then, we might have a need for hyperpnea from circulatory causes. As a matter of fact our own studies of the blood gases in pneumonia rarely show evidence of stagnation of blood flow.

A fourth cause for hyperpnea is anoxemia, or deficient oxygen saturation of the blood as it passes through the lungs. Such a condition is frequently found in pneumonia and may result from the admixture of aërated and unaërated blood or from an imperfect aëration of the entire blood mass as it passes through the lungs. The former might result from consolidation or localized edema, the latter from generalized edema or from the presence of exudate or foam in the air passages.

How anoxemia causes hyperpnea is a disputed matter, and for the purpose of the present analysis it is not necessary that we go

* In the matter of the P_h of the blood the normal can be taken as 7.35. How much variation there is in health is unknown. Several of our cases show P_h of 7.25 or less, which we believe is lower than the normal range of variation. Our figures have all been corrected for oxygen unsaturation by Peters's formula.[14]

into it. The normal stimulus to the respiratory center is the reaction of the blood. Anoxemia may effect it through the production of asphyxia acidosis.

There are one or two other factors which might play a part in the determination of the required ventilation in pneumonia. For one thing there is the matter of the dead air space in breathing. In the normal person alveolar and total ventilation run roughly parallel, for ventilation increase is met by both increase in rate and depth. It can be seen, however, as we have said earlier, that unless there was a compensatory change in the size of the dead space, with a rapid shallow type of breathing, it would take a relatively greater total ventilation to secure a given alveolar ventilation than with a slow deep type. The rapid shallow type is, as has been pointed out, a less economic type; a greater total effort is required to secure a given result.

An enlargement in the dead space would also require a greater total ventilation to secure a given alveolar ventilation. Such an enlargement occurs in emphysema, as has been shown by Hoover.[19] So far as we know it has not been studied in pneumonia. In the emphysematous patient with pneumonia, however, it might be expected to be still another factor demanding hyperpnea.

So much for the factors which may play a part in the determination of the pulmonary ventilation required of the pneumonia patient. Any or all of them, if present; will exert their influence in the direction of demanding a greater ventilation than the person would have under normal circumstances.

Let us now turn to the matter of supply, to the ventilatory powers of the pneumonia patient in contrast to his ventilatory needs. This in the last analysis boils down to the vital capacity and to the type of his breathing. The lower the vital capacity the more will a patient have to increase his ventilation by an increase in rate at the expense of depth. That the vital capacity is reduced in pneumonia is certain. A few observations that we have made show there may be a reduction to one-third or less of the normal. This may be due to actual obliteration of air space by the pathologic process or it may be due to inability to take a deep breath because of pleural pain or to interference with

proper action of the diaphragm due to abdominal distention. Whatever the cause it will have the effect of necessitating a rapid shallow type of breathing.

In regard to such a type of breathing it has been shown by Meakins[2] actually to exist in pneumonia, and it has been shown by Haldane, Meakins and Priestley[20] to be of itself capable of producing anoxemia. This they think is due to an incomplete expansion of the lung in very shallow breathing and the admixture of poorly aërated blood from poorly ventilated portions with properly aërated blood from properly ventilated portions of the lung.

To summarize the morbid physiology of pneumonia, then, we may say that the pneumonia patient may be called upon to ventilate his lungs more than he would under normal circumstance for any or all of the following causes:

1. Increased metabolism due to fever.
2. Difficulty in eliminating CO_2 with resulting carbonic acidosis.
3. Deficient circulation rate or blood flow.
4. Anoxemia.

His demand for ventilation, in other words, is increased. Now at the same time his available supply of ventilation dependent on his vital capacity may be markedly decreased. His respiratory mechanism is confronted simultaneously with more work to do and a lessened ability for doing it. Not only that, but because of the low vital capacity he is obliged to meet his demand for increased ventilation by adopting a rapid shallow type of breathing which of itself introduces two vicious circles, the first that, being an uneconomic type of breathing, it still further increases the necessary total ventilation, and the second that this type has been shown to increase the degree of anoxemia. It would seem that we need look no further for an explanation of the dyspnea. Moreover, the multiplicity of the factors involved and the fact that various combinations of them may serve to use up all respiratory reserve will show why such clinical phenomena as dyspnea and cyanosis bear no definite relation to the extent of the anatomic process in the lungs. With this analysis of the morbid physiology in mind let us try to deduct the logical indications for symptomatic treatment in pneumonia. The possible lines to pursue would seem to be as follows:

1. Procedures which diminish the need for ventilation.
2. Procedures which increase the vital capacity.

In other words, measures which will either decrease demand or increase supply of ventilation.

As to the first, we have several methods of attack. The metabolism we probably could only alter by altering the temperature. Hyperpyrexia we do try to relieve, but it is not felt wise nowadays to interfere with temperature unless excessive. At the crisis Nature restores temperature to its normal level. The metabolism probably falls with it, and, as is well known, dyspnea often ceases with no discoverable change in the extent of the anatomic lesion. This may well be explained by a suddenly decreased demand for ventilation.

Deficient blood flow we may prevent or relieve by suitable cardiac stimulation.

In the matter of increasing vital capacity, and so increasing the efficiency of the pulmonary bellows, there probably are no direct measures that can be used except when the low capacity is due to pleural pain, in which case morphin may accomplish it, or when it is due to abdominal distention, when relief of that condition may improve it.

There are, however, two procedures which meet direct indications which we wish particularly to discuss; they both are of the type which may be expected to diminish the need for ventilation. One is the administration of alkali and the other the therapeutic administration of oxygen.

In regard to alkali we have found, as has been stated earlier, that the CO_2 dissociation curve is usually at a normal or nearly normal level. This means there is either a normal or slightly reduced amount of available alkali in the blood. Our findings in this respect are in agreement with those of Palmer.[21] This writer, however, has found there is a considerable quantity of organic acid eliminated in the urine of pneumonia patients. It has also been found by Palmer[22] and others that it takes more alkali by mouth to render the urine alkaline in pneumonia than in the normal person. In considering the preservation of acid base equilibrium in disease we must bear in mind there are two principal compensatory

mechanisms, the pulmonary and the renal. The level of the dissociation curve indicates that the renal compensation is either not at all or only slightly disturbed. The organic acid found in the urine by Palmer must be eliminated by the kidney nearly as fast as produced, for did it accumulate in the blood the dissociation curve would be definitely lowered. The pulmonary response is frequently adequate also, as shown by the normal P_h of the blood, although, as we have said before, in certain severe cases there is apparently a shift in reaction in the acid direction, with a curve at a normal level, which we can only interpret as carbonic acidosis due to insufficient pulmonary compensation.

If there is either no reduction or only slight reduction in the bicarbonate content of the blood, as shown by the level of the dissociation curve, why is it that the alkali tolerance is increased? This may be in part because, owing to a urinary acidity increased by the presence of organic acid, it actually takes more alkali to change the reaction of the urine itself, but in greater part it seems to us that it may be due to a retention of alkali as a compensatory measure on the part of the body.

As Y. Henderson and Haggard have shown, to eliminate a given quantity of CO_2 per minute less pulmonary ventilation will be required at a high level of blood bicarbonate than at a low one. That is to say, with a high dissociation curve less ventilation will care for a given CO_2 output than with a low curve. A raising of the curve, therefore, enables the respiratory mechanism to perform a required task with less labor. Scott[23] has shown that patients with pulmonary emphysema do this very thing. They have a greatly reduced ventilating capacity; they compensate by raising their dissociation curve.

Now it occurs to us that this same sparing of the respiratory mechanism may occur in pneumonia as a result of giving sodium bicarbonate. When the curve is below the normal zone this, of course, will be beneficial; but even when it is at a normal level to start with, on giving alkali it goes to a higher level than normal. This in a patient with a normal P_h before alkali might theoretically, at least, reduce the amount of pulmonary ventilation that he would have to produce, and in one with a low P_h a raising of the

curve might change an existing ventilation from an insufficient to a sufficient one. Some of our cases seem to show that even without the therapeutic administration of alkali Nature attempts to raise the curve, for after the crisis it was at a considerably higher level

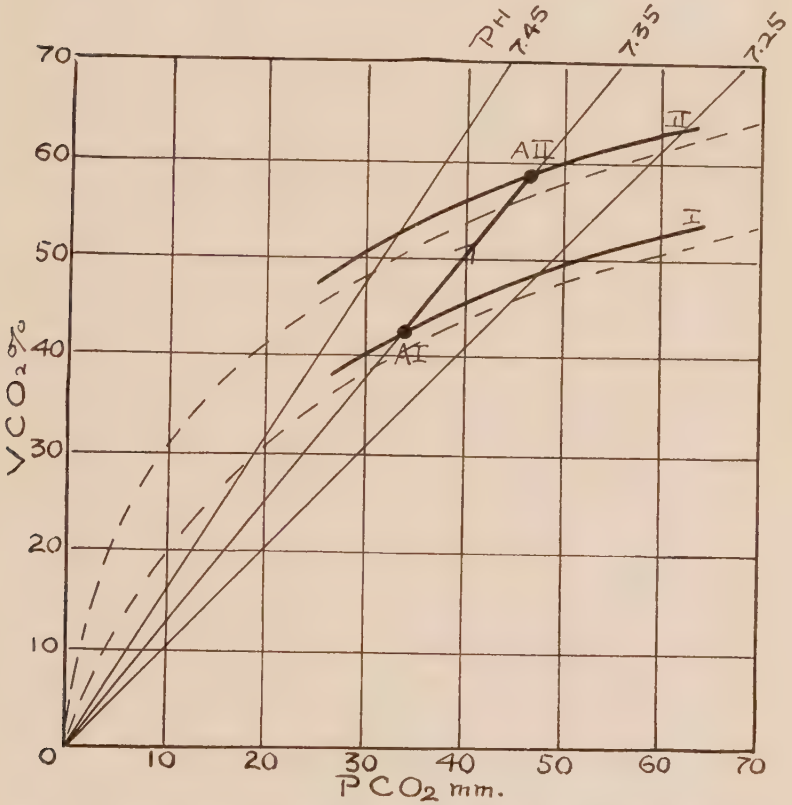

FIG. 1.—Diagram showing that in a pneumonia patient with a dissociation curve at a normal level (Curve I), that is to say falling within the normal zone (indicated by interrupted line) and a normal P_h of 7.35, as shown by the position of the arterial point *AI*, the effect of raising the curve to the position of Curve II by the administration of bicarbonate would be to decrease the volume of pulmonary ventilation necessary, for with the arterial point in the position *AII* the CO_2 tension of the arterial blood will be 46.5 mm.; when it was in the position *AI*, however, it had to be kept down to 34 mm. With an unchanged CO_2 output considerably less ventilation will be needed to keep the arterial CO_2 tension at 46.5 mm. than at 34 mm. The effect of alkali then would be to diminish the task of the pulmonary bellows. Figs. 1 and 2 are hypothetical, but are in many respects similar to actual curves which we shall report later.

Note.—VCO_2 = CO_2 content of blood in volume per cent. PCO_2 = CO_2 pressure of blood in millimeters of Hg.

than before. We have also found that the curve can be readily raised 10 volume per cent or more by giving alkali.

Our conception, then, of the use of alkali in pneumonia is that it may be helpful by correcting a non-volatile acidosis when that exists, and even when it does not, by decreasing the amount of ventilation necessary to accomplish the output of a given quantity of CO_2, or in the cases which show a lowering of the P_h by rendering an ineffective ventilation effective. These actions are shown hypothetically in Figs. 1 and 2. In fact, alkali given therapeutically we believe may act as a conservator of respiratory effort. The actual effect of alkali has been studied in a very few cases. The finding of the indication, however, we believe is definite and the use of alkali worthy of further study. That it is desirable to diminish respiratory effort we believe is obvious, but pertinent in this regard are the findings of Newburgh, Means and Porter,[24] which showed that in experimental pneumonia as the disease advanced there was a progressive decrease in the sensitivity of the respiratory center ending finally in death from respiratory failure. It has also been shown more recently by Davies, Haldane and Priestley[25] that resistance to respiration in time fatigues the center. It is entirely conceivable that the abnormal respiratory load in patients with pneumonia has this same effect, and that removing or decreasing this load might greatly improve the chance of recovery. A word of warning, however, about the use of alkali is in order. It was shown in our previous paper,[15] and has been shown by others, that an alkalosis can be produced by overdosage with sodium bicarbonate. To our minds, following Palmer,[22] the reaction of the urine should be the guide. Enough bicarbonate should be given to make the urine alkaline; then it should be discontinued, to be recontinued later if urine returns to an acid reaction. This method we believe safe. It is inconceivable that a dangerous state of alkalosis could develop while the urine remained acid, for the response to alkalosis by the body would be the elimination of the base by the kidney.

The second therapeutic measure that we have studied in detail is the use of oxygen. Stadie[26] and Meakins[27] have both found that there often is an arterial or anoxic[28] anoxemia in pneumonia. We

have had the same experience. In this clinic in the past year there have been treated by oxygen inhalation ten patients with lobar

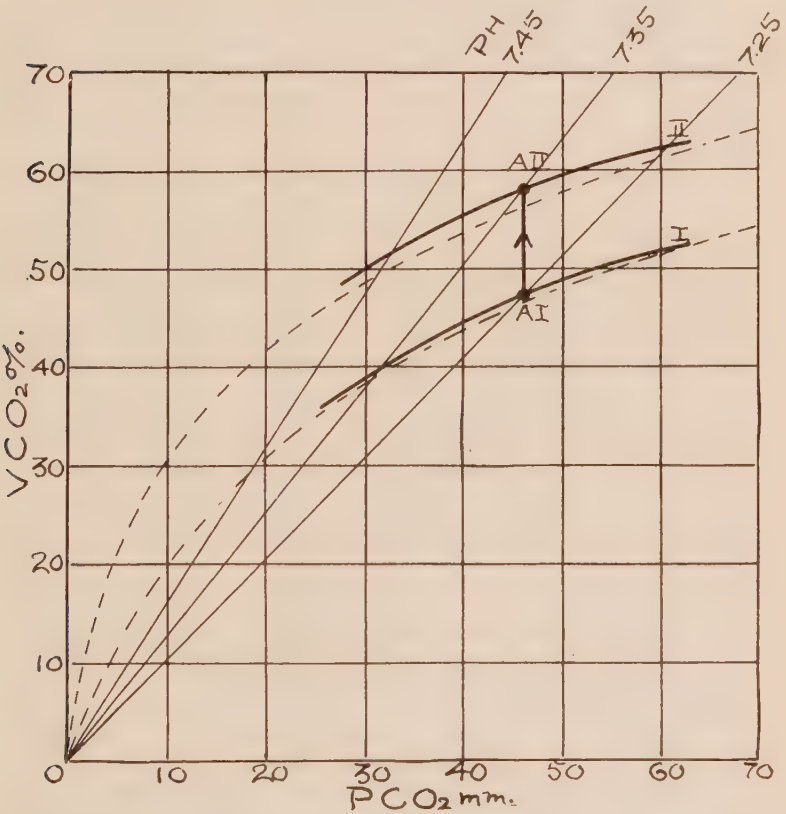

FIG. 2.—Diagram showing that in a pneumonia patient with a P_h shifted in the acid direction, as shown by the position of the arterial point *AI* on the P_h 7.25 diagonal, the effect of raising the curve from the position *I* to *II* will be to convert an insufficient ventilatory response into a sufficient one. The same pulmonary ventilation will be needed to keep the arterial point in the position *AII* when the curve is in the position *II*, as was required to keep it at *AI* when the curve was in the position *I*. But in the position *I* the P_h is 7.25, which is acidotic, while in the position *II* it is 7.35 or normal. The effect of alkali has been to restore pulmonary compensation with no added effort on the part of the ventilatory mechanism. Elevation in the curve by alkali of the order of magnitude shown in this and in Fig. 1 has been found quite easy of accomplishment.

and two with bronchopneumonia. All had arterial anoxemia in some stage of the disease, except one of the cases of bronchopneu-

monia. The arterial saturation in the majority of these was from 74 to 92 per cent; the lowest observed was 62.3 per cent. Of the ten lobar pneumonia patients there were eight in whom blood-gas determinations were made in relation to the oxygen therapy. The arterial saturation was materially raised in all but one by

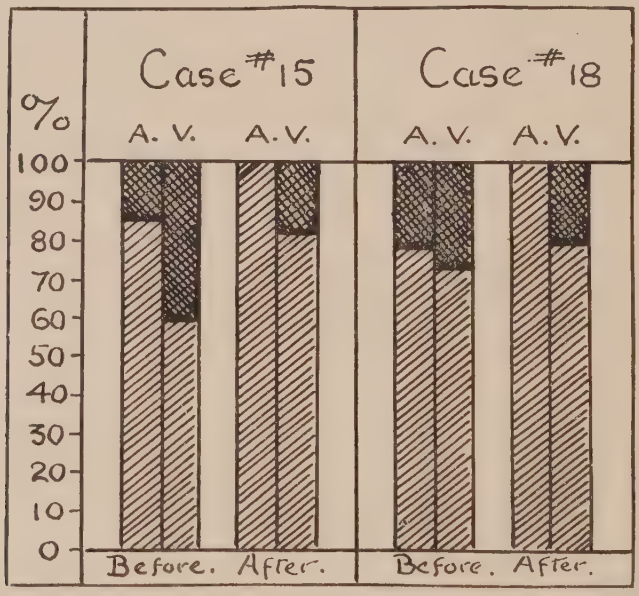

FIG. 3.—Showing the effect of oxygen therapy on the arterial and venous oxygen saturation in two cases of lobar pneumonia. *A*, arterial blood; *V*, venous blood. The height of the lightly shaded portion of the column indicates the percentage oxygen saturation of the given blood. The observations in Case 15 were just before and immediately after twenty minutes of oxygen therapy. It will be seen that a moderately severe arterial anoxemia was entirely abolished. The observations in Case 18 were before and after oxygen had been given intermittently for a total of thirteen out of twenty-four hours. Here a very severe arterial anoxemia was abolished.

oxygen administration when a suitable apparatus was employed. In four it was raised to the normal level. In the case of broncho-pneumonia, which showed arterial anoxemia, the saturation also increased after oxygen.

Objectively the effect of oxygen therapy was nearly always to lessen or abolish cyanosis, and also to slow very definitely the

pulse-rate anywhere from 5 to 20 beats. Subjectively there sometimes was relief to respiratory distress and sometimes not.*

It seems to us that the presence of anoxemia constitutes a perfectly definite indication for treatment. Anoxemia is, for one thing, one of the possible causes of hyperpnea and therefore its relief may be expected, to a certain extent, to reduce the respiratory burden; in addition it is the cause of a number of very unpleasant symptoms *per se*. Barcroft,[29] for example, when in an anoxemic state produced in a chamber with rarefied air suffered from distressing headache, nausea, vomiting, faintness and visual disturbances. His degree of anoxemia was not as great as is often found in pneumonia. In severe anoxemia profound damage may be done to the cardiovascular and central nervous systems. It would seem, then, that the relief of anoxemia might spare a patient from a multiplicity of injurious effects. The slowing of the pulse-rate when anoxemia is relieved would indicate an improved heart action. It further has been shown, both by the work of Meakins and of ourselves, that anoxemia in pneumonia can usually be decreased or abolished by suitably conducted oxygen therapy.[27] The results in two of our treated cases are shown graphically in Fig. 3. For the purpose, oxygen may often have to be given continuously or at frequently repeated intervals. Some of the more modern types of apparatus will have to be used and in most instances a special nurse will be necessary to carry out the treatment.

Summary. 1. Rational symptomatic treatment must be based on a clear understanding not only of the morbid anatomy but the morbid physiology as well of the disease concerned.

2. In pneumonia the outstanding feature is the respiratory battle. A great strain is thrown upon both respiratory and circulatory mechanisms.

3. The respiratory strain results from the circumstance that the pneumonia patient is simultaneously confronted with a necessity for a greater pulmonary ventilation than under normal circumstances, and a pulmonary bellows of reduced efficiency with which

* The data on oxygen therapy will be published shortly in full by Barach and Woodwell.

to accomplish it. He has a greater ventilatory demand and at the same time a decreased ventilatory supply.

4. A number of factors contribute to this vicious state of affairs. Increased metabolism, acidosis, deficient circulation and anoxemia may increase the demand for ventilation. At the same time decreased vital capacity due to consolidation, edema or pleural pain or abdominal distention may decrease the capacity of the pulmonary bellows.

5. We can directly meet some of these indications. Acidosis if present we can correct by alkali administration. The type of acidosis when present is often a carbonic acidosis. Alkali administration also may be hoped to help the non-acidotic case because it raises the bicarbonate level of the blood, which in turn makes it possible for the patient to get along with a reduced pulmonary ventilation.

6. Anoxemia, which is frequently present and which has a variety of injurious effects, may be corrected or relieved by oxygen administration.

7. Both these measures—alkali administration and oxygen administration—must be carefully and intelligently controlled. Bicarbonate should be given only in amounts sufficient to turn the urine alkaline to litmus. If pushed further than this it may do harm by producing alkalosis. Oxygen should be given with one of the modern types of apparatus and often nearly continuously by a specially instructed nurse. Its continuation is to be governed by the effect on the cyanosis and the comfort of the patient.

8. These measures are supplementary to specific therapy. When used, however, they may be expected to spare the patient several avoidable burdens and leave him free to devote his entire energy to the fighting of his infection, thus theoretically at least improving his chance of recovery.

BIBLIOGRAPHY

1. Peabody, F. W., et al: Clinical Studies on the Respiration, I, III and IV, Arch. Int. Med., 1915, xvi, 846; ibid., 1917, xx, 433; ibid., 1917, xx, 443.

2. Meakins, J. C.: Harmful Effects of Shallow Breathing, with Special Reference to Pneumonia, Arch. Int. Med., 1920, xxv, 1.

3. Edsall, D. L.: Clinical Study of Respiration, Boston Med. and Surg. Jour., 1912, clxvii, 639.

4. Boothby, W. M.: Determination of the Circulation Rate in Man at Rest and at Work, Am. Jour. Physiol., 1915, xxxvii, 383.

5. Means, J. H., and Newburgh, L. H.: Effect of Caffein on the Blood Flow in Normal Human Subjects, Jour. Pharm. and Exper. Therap., 1915, vii, 449.

6. Peabody, F. W.: Vital Capacity of the Lungs and its Relation to Dyspnea, Arch. Int. Med., 1917, xx, 443.

7. Grabfield, G. P., and Means, J. H.: Effect of Caffein on the Reaction to CO_2 of the Normal Human Respiratory Mechanism, Jour. Pharm. and Exp. Therap., 1917, x, 159.

8. Means, J. H., and Balboni, G. M.: Various Factors of Respiration in Persons with Pneumothorax, Jour. Exp. Med., 1916, xxiv, 671.

9. Coleman, W., and Du Bois, E. F.: Clinical Calorimetry VII, Arch. Int. Med., 1915, xv, 887.

10. Barr, D. P., and Du Bois, E. F.: Clinical Calorimetry XXVIII, Arch. Int. Med., 1918, xxi, 627.

11. McCann, W. S., and Barr, D. P.: Clinical Calorimetry XXIX, Arch. Int. Med., 1920, xxvi, 663.

12. Haggard, H. W., and Henderson, Y.: Hematorespiratory Functions, Jour. Biol. Chem., 1919, xxxix, 163.

13. Henderson, L. J.: Das Gleichgewicht zwischen Basen and Säuren im tierischen Organismus, Ergebn. Physiol., 1909, viii, 254.

14. Peters, J. P., Jr., Barr, D. P., and Rule, F. D.: CO_2 Absorption Curve and CO_2 Tension of the Blood of Normal Resting Individuals, Jour. Biol. Chem., 1921, xlv, 489.

15. Means, J. H., Bock, A. V., and Woodwell, M. N.: Studies of the Acid-base Equilibrium in Disease from the Point of View of Blood Gases, Jour. Exp. Med., 1921, xxxiii, 201.

16. Peters, J. P., Jr., and Barr, D. P.: CO_2 Absorption Curve and CO_2 Tension of the Blood in Cardiac Dyspnea, Jour. Biol. Chem., 1921, xlv, 537.

17. Hoover, C. F.: Moisture in the Air Spaces of the Lungs and Oxygen Therapy, Jour. Am. Med. Assn., 1918, lxxi, 880.

18. Pearce, R. G.: The Cardiorespiratory Mechanism in Health and Disease, Arch Int. Med., 1921, xxvii, 139.

19. Hoover, C. F.: Alveolar Air and Minute Volume of Air in Pulmonary Emphysema, Tr. Assn. Am. Phys.,

20. Haldane, J. S., Meakins, J., and Priestley, J. G.: Harmful effects of Shallow Breathing, Jour. Physiol., 1919, lii, 433.

21. Palmer, W. W.: Acidosis and Acid Excretion in Pneumonia, Jour. Exp. Med., 1917, xxvi, 495.

22. Palmer, W. W., and Henderson, L. J.: Clinical Studies of Acid-base Equilibrium and Nature of Acidosis, Arch. Int. Med., 1913, xii, 153.

23. Scott, R. W.: Observations on the Pathologic Physiology of Chronic Pulmonary Emphysema, Arch. Int. Med., 1920, xxvi, 544.

24. Newburgh, L. H., Means, J. H., and Porter, W. T.: Respiratory Mechanism in Pneumonia, Jour. Exp. Med., 1916, xxiv, 583.

25. Davies, H. W., Haldane, J. S., and Priestley, J. G.: The Response to Respiratory Resistance, Jour. Physiol., 1920, liii, 60.

26. Stadie, W. C.: Oxygen of Arterial and Venous Blood in Pneumonia and Its Relation to Cyanosis, Jour. Exp. Med., 1919, xxx, 215.

27. Meakins, J.: Observations on the Gases in Human Arterial Blood in Certain Pathological Conditions and Their Treatment with Oxygen, Jour. Path. and Bact., 1921, xxiv, 79.

28. Barcroft, J.: Anoxemia, Lancet, 1920, cxcix, 485.

29. Barcroft, J., Cooke, A., Hartridge, H., Parsons, T. R., and Parsons, W.: The Flow of Oxygen Through the Pulmonary Epithelium, Jour. Physiol., 1920, liii, 450.

DISCUSSION

Dr. W. C. Stadie, New Haven: The tension at which oxygen is supplied to the tissues is equally if not more important than the total quantity in tissue respiration. In anoxemia the blood is only partially saturated and hence delivers oxygen to the tissues at low tensions. Krogh has shown that the partial pressure of oxygen in the tissues is practically the same as that of venous blood, contrary to the hitherto accepted idea that it is zero. It is probable that the tissues do not function well if oxygen is delivered to them at low tensions. This is particularly true of highly specialized tissue such as the central nervous system. Unless this were so it would be difficult to explain the harmful effects of anoxemia. The extreme distress exhibited when the blood is 30 to 40 per cent unsaturated is in striking contrast to the abundance of oxygen still present (12 to 14 volumes per cent), but this oxygen is available only at tensions below 35 mm. of Hg instead of 110 mm.

Anoxemia is a regular accompaniment of pneumonia and often becomes quite marked. We have found that severe anoxemia is associated with the fatal cases, and, as a rule, cases which develop an oxygen unsaturation greater than 20 per cent die. The anoxemia is relieved by the inhalation of oxygen. In order that oxygen treatment be of any value it is essential that the treatment be continued over a long period of time, the percentage of oxygen be fixed at any desired level, discomfort and inconvenience of masks, funnels and tubes eliminated and adequate medical and nursing attention rendered at all times. This we have accomplished by means of a specially constructed oxygen chamber in which, so far, we have treated eight cases. Controlled studies of the blood and respiration have been done on these patients, of which a detailed report will be published later. The results on this series is encouraging and seem to show that oxygen properly administered is a valuable therapeutic measure in the treatment of the anoxemia of pneumonia.

Dr. G. Kober, Washington, D. C.: I have greatly enjoyed Dr. Means's excellent contribution to the symptomatic treatment of pneumonia. I desire to recall the so-called eliminative treatment advocated by Hugh Bennett in his classical lectures over fifty years ago. This treatment consisted in the administration of tablespoonful doses of a mixture composed of liquor ammon. acet., fℨvij, and spts. ætheris nitros, fℨj, repeated every two or three hours until diaphoresis set in, then the intervals were lengthened. In the light of modern pathology these diaphoretic and diuretic agents evidently subserve a useful purpose in the elimination of toxins, and the blood being determined more or less to the periphery may also have a beneficial effect on the heart and lungs. I have resorted to this treat-

ment ever since I observed its good effects at the Carlisle Cavalry Recruiting Depot in 1868, and cannot endorse it too highly. It appears to prevent the development in all but bad chronic heart cases of the dangerous symptoms such as demand the administration of oxygen.

Dr. C. F. Hoover, Cleveland, Ohio: Air hunger and cyanosis are not necessarily companion symptoms in lobar pneumonia. Be it understood that air hunger is a purely subjective symptom, and means that respiratory need is brought within the field of the patient's consciousness, also that by cyanosis in this relation is meant oxygen unsaturation of the aortic blood. Air hunger due to the amount of lung consolidation in pneumonia is unusual. Hyperpnea of a degree tolerated with comfort is the rule, but it is caused by fever and not by anoxemia or lung consolidation. When air hunger does occur in pneumonia it is usually due either to moisture in the air spaces from inflammatory edema or to cardiac incompetence. There may be demonstrable enlargement of the two ventricles on account of the increase minute volume flow of blood, just as we see this symptom occur in Graves's disease and in arteriovenous fistula; but when cardiac incompetence supervenes, ectasis of the right auricle is always demonstrable.

If in the clinical course of this disease cyanosis is present without air hunger, the arterial blood is anoxemic but does not reveal a reciprocal rise in its carbon dioxide content, and at autopsy the pneumonic area contains a variable but always a considerable amount of blood.

Although very convincing, these facts are thus far the only evidence for the contribution of unrespired blood to the aortic stream from a pneumonic area. If as much as one lobe of the lung is consolidated and the arterial blood has no oxygen unsaturation (as occurs in tuberculous pneumonia) the autopsy will then reveal an ischemia of the consolidated area. In such cases there is no clinical evidence for the contribution of unrespired blood to the aortic stream.

Digitalis therapy is of service only when there is demonstrable dilatation of both ventricles and the right auricle.

Dr. S. Solis-Cohen, Philadelphia: The careful and scientific studies which have been presented bear out the clinical experience of many years. One physician at Atlantic City uses sodium bicarbonate as a specific in pneumonia. While not going that far, personal observation has shown me the value of maintaining a constant alkalinity of the urine. A nurse, or an intelligent member of the family, may be instructed to make a test with litmus paper at each voiding and to regulate the dosage of alkali accordingly. As a scientific rule this is open to criticism; as a rule of thumb, workable in the sick room, it answers a good purpose. One may use sodium bicarbonate alone or in conjunction with any alkaline diuretic that he prefers. Sometimes the alkaline diuretic is sufficient in itself. Alkalized blood is more

readily oxygenated. The empiric use of oxygen has gone through many successive waves of acceptance and rejection. Oxygen will not, indeed, revive the dead; but if the patient really gets it, and gets it in time, it is life-saving. It must, however, be administered systematically, and in proper fashion. Merely to let it play about the nose of the patient, or to use it irregularly or spasmodically, is futile. If we give it effectively by continuous administration and oral inhalation we may, perhaps, waste a lot of the gas, but we will save many lives. Another matter worth comment is abdominal distention. Physicians do not always realize the frequency and seriousness of dilatation of the stomach. In the old days when we saw a good deal of typhoid we percussed the right chest as a routine to detect the earliest obscuration of liver dulness; in pneumonia we must percuss the left side to detect the earliest sign of distention of the stomach, which is much more frequent than intestinal distention only, and is very likely to have fatal consequences if not promptly relieved. Administration of the posterior pituitary principle, about every third hour, but guided by the systolic blood-pressure, is an integral part of what I have termed the "definite" treatment of pneumonia. This, of itself, tends to prevent gastric and intestinal distention. Should gastrectasia occur nevertheless—and as soon as detected through tympany extending beyond the midaxillary line on the left, or as soon as suggested by a tendency to hiccough—the stomach is to be washed out and 1 cc of the strong posterior pituitary preparation injected intramuscularly every hour for three hours; after which, if need be, eserine may be used in substitution for the pituitary principle or in alternation with it. Usually the lavage and the three injections of the pituitary solution suffice for the time being.

THE BEARING OF THE EVOLUTION OF THE PNEUMONIC EXUDATE ON LOCAL CHEMICAL FACTORS INFLUENCING RECOVERY AND RESOLUTION IN LOBAR PNEUMONIA

By FREDERICK T. LORD, M.D.
BOSTON, MASS.

In considering local chemical factors influencing recovery in lobar pneumonia the importance of humoral factors should not be disregarded. The demonstration of protective substances in the blood serum of pneumonic patients at or about the time of crisis and the curative action of Type I antipneumococcus serum indicate a humoral aspect to recovery. Such factors may be regarded as important in checking septicemia and limiting the spread of the local lesion. But among other matters the successive invasion of different lobes, the recession in one in the face of accession in another lobe, the frequency of relapsing pneumonia and the ready growth of pneumococci in pneumonic and postpneumonic blood serum are difficult of comprehension on the basis of humoral factors as a sole explanation, and suggest that other factors may also be concerned in recovery.

Recovery may be influenced by such local factors as (*a*) partial isolation of the pneumonic exudate, separated from the general circulation by the alveolar wall, and a restricted circulation through the involved area, favoring chemical changes little reflected in the body as a whole; (*b*) the evolution of the pneumonic exudate through an early period of engorgement with serum to a later stage of cellular exudation, and (*c*) a local increase in H-ion concentration.

In the theory of local factors in recovery the sensitiveness of the pneumococcus to increase of acidity and the influence of varying

concentrations of serum on acid death-point are significant. Thus in mixtures of buffered isotonic bouillon and varying concentrations of serum the greater the H-ion concentration within certain limits the more rapid the death of the pneumococcus. For example, at pH 6.1 the viability of the pneumococcus in bouillon alone is about four hours while the addition of 1 per cent or more of serum at the same pH prolongs the life of the culture for many days; but in less than 1 per cent serum at this pH the organism lives for only a few days. At pH 5.5 the viability in bouillon alone is about two hours, and even 100 per cent serum may not suffice to prolong the life of the culture for more than twenty-four hours. In explanation of recovery it may be conceived that as the evolution of the exudate takes place with an increasing local H-ion concentration and diminishing amount of serum the acid death-point of the pneumococcus is reached and crisis follows.

Resolution may be ascribed to (*a*) autolysis, the polynuclear leukocytes being the chief source of ferment; (*b*) an increase in local H-ion concentration, and (*c*) ferment-antiferment balance. As previously shown the pneumonic exudate contains two enzymes: One capable of digesting coagulated albumin at pH 7.3 to 6.7 and another capable of splitting peptone to amino-acid nitrogen at pH 6.3 or 5.2. R. N. Nye and I have shown that in the balance between pneumonic cellular material (enzyme) and serum (antienzyme) enzymatic action is absent with more and present with less than about three parts serum to one part cells. In the evolution of the exudate as the amount of serum diminishes and the number of cells and the H-ion concentration increase, conditions become suitable within the alveoli for the digestive action of the enzyme and resolution follows. The usual restoration of the lung as a whole to normal is possible by the presence of serum in the pulmonary capillaries protecting the lung framework from enzymatic action. An excess of serum within the alveoli or a deficit of enzyme may lead to delayed resolution. Postpneumonic abscess may follow an excess of ferment or a deficit of serum.

DISCUSSION

Dr. Torald Sollmann, Cleveland, Ohio: Is there any evidence that this appears to take place during life or is it a postmortem change?

Dr. F. T. Lord (closing): The determination of the H-ion concentration of the pneumonic lung was made post mortem in man and dogs. In experimental pneumonia in dogs the chest was opened, the lung removed while the animal was under an anesthetic and while the heart was still beating and the H-ion concentration at once determined. An increase in the H-ion concentration to within a range (pH 6 to 5.4), at which the viability of the pneumococcus is shortened, was thus demonstrated. There are several sources of possible error. Postmortem increase in H-ion concentration cannot be excluded. The determinations were made on the fresh juice of the involved part of the lung. Admixture of blood from the blood-vessels would tend to lower rather than increase the H-ion concentration of the mixture. It seems impossible by any available method to determine during life the H-ion concentration of the exudate alone.

TRACHEAL AND BRONCHIAL STENOSIS AS CAUSES FOR EMPHYSEMA

By C. F. HOOVER, M.D.
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During the past ten years much research has been done on the subject of emphysema and asthma and on the chemistry of blood and respired air, but few studies on the mechanism of bronchiolar spasm and emphysema have been published. Some of the most recent work on this subject has been based on the assumption that the time-honored teaching of the causal relation between expiratory dyspnea and emphysema and asthma is unassailable. To the writer it seems the prevailing teaching on these subjects needs revision. To learn the mechanism of emphysema and asthma by way of the chemistry of the blood gases and of the respired air seems quite hopeless. When a clear understanding of the mechanism of emphysema and asthma has been attained we shall be in a much stronger position to interpret our chemical studies of the blood gases and respired air.

Biermer's Theory. The modern interpretation of bronchiolar asthma originated with Biermer, of Zurich, who in 1870 published an exposition of the subject in *Volkmann's Sammlung klinischer Vorträge*.[1] Biermer referred to Paul Bert as having finally proved that vagus excitation causes bronchiolar contraction, although it had been suspected and claimed in fact by Williams in 1840 at a meeting in Glasgow (the tenth meeting of the Association for the Advancement of Science). Biermer said the low position of the diaphragm could not be reconciled by his contemporaries to bronchiolar spasm as an explanation for pulmonary emphysema. Bam-

[1] Innere Medicin, Leipzig, 1870–75, No. 3.

berger had believed phrenic spasm to be the source of acute pulmonary emphysema, and Lehman took the same view.

It is with much satisfaction that Biermer quotes the experiments of Paul Bert, which were published in 1870 in his book on *The Comparative Physiology of Respiration*. Biermer conceived an essential difference to exist between stenosis in the remote branches of the bronchi and stenosis in the trachea. He regarded dyspnea of bronchiolar stenosis as essentially expiratory, because he believed the active character of the expiration of asthma produced a vicious cycle; an active pressure on the lung compressed the boundaries of the respiratory units; the air cells were subjected to compression, and the air passages connecting them with the larger bronchi were also compressed—that is, the more active the expiratory effort the greater grew the resistance to exit of air from the respiratory units. On this account the expiratory phase was supposedly much prolonged. Biermer says the difference between tracheal and bronchiolar stenosis lies in the fact that the former is an inspiratory and the latter an expiratory dyspnea. As an evidence of this point of view he calls attention to the inspiratory retraction in the supraclavicular, suprasternal and epigastric regions in tracheal stenosis and contrasts it with the labored expiration of asthma; and he calls attention to the want of evidence of active vigor in the expiratory phase in cases of tracheal stenosis.

Nature of Acute Emphysema. By way of introduction to the subject, let it be understood that we are dealing with two phenomena, viz., bronchiolar hypertonus and emphysema of the lungs, which consists solely of increase in the pulmonary residual air. In such cases there is only altered physiologic function and no anatomic disease of the lung.

Bronchiolar spasm is immediately attended with emphysema, diminution of vital capacity, inspiratory and expiratory dyspnea and lessened extensibility and compressibility of the lungs. Thus far we have no evidence that disturbances in the pulmonary circulation contribute to rigidity of the lungs in acute pulmonary emphysema. The loss in extensibility is due entirely to emphysema and narrowing of the bronchial tree. Although we have no positive evidence for hyperemia of the pulmonary circulation in bronchiolar

spasm there is negative evidence against its responsibility for rigidity of the lung. That hyperemia can reduce the vital capacity of the lung quite as much as does bronchiolar spasm has been abundantly observed. Both conditions may reduce the vital capacity of the lung to 800 cc in persons who normally may have a vital capacity of 4000 cc. It is, however, incomprehensible how such a hyperemia could occur without causing great resistance to the work of the right heart and thus causing ectasis of the right ventricle and right auricle. I have never seen any evidence of enlargement of the right side of the heart in acute bronchiolar spasm; evidence for it has never been adduced, although in literature on the subject this assumption commonly appears. The mechanism of bronchiolar spasm, with all its attending attributes above described, may appear and disappear with such suddenness that it seems inconceivable that the collective phenomena can have any other origin than a neuromuscular performance.

This aspect of acute emphysema is best seen in patients who have periodical bronchiolar spasm from vagus excitation due to mediastinitis. Such a patient came under my observation during an attack. The patient was a little girl, eight years old, who for the last two years had been quite well, according to the grandmother's account, excepting for asthma, which occurred every few weeks and lasted only half an hour. The last attack of asthma this child had suddenly terminated while I was observing the volume and excursion of her lungs. The lungs filled the pleural sinuses and the child was employing her utmost effort to breathe during both inspiration and expiration. Each respiratory cycle exhibited her vital capacity, when suddenly, in the midst of a single respiration, the lower border of the lung was seen to ascend 4 cm., and with this rapid disappearance of emphysema there was a complete return of respiratory comfort. It can be truthfully said that this child began a respiratory cycle with emphysema, lessened vital capacity, inspiratory and expiratory dyspnea, lessened extensibility and compressibility of her lungs, and ended the cycle with respiratory euphoria. It is inconceivable that such a rapid disappearance of all these symptoms could be produced by any other than a neuromuscular phenomenon. The recovery was too rapid to include vasomotor phenomena, with

consequent changes of blood distribution, as among the causes of the symptom-complex. We have therefore very good reasons for excluding all proposed possibilities excepting bronchiolar spasm as the cause of acute emphysema. But we find the theory of the mechanism by which bronchiolar spasm is supposed to produce emphysema and its attendant symptoms does not withstand clinical and experimental criticism.

Tracheal Stenosis. Biermer's theory teaches that bronchiolar spasm is attended with an expiratory effort which produces a vicious cycle by compressing the bronchiolar exits from the respiratory units. If this be true a resistance anywhere in the tracheobronchial tree which is sufficient to demand expiratory compression of the lung should lessen the volume flow toward the latter part of expiration, as the intrapleural pressure rises, and result in emphysema and prolongation of the expiratory phase. If a strong muscular effort is needed to accomplish expiration against tracheal resistance the same mechanism of expiratory stenosis should be operative that Biermer's theory teaches for bronchiolar hypertonus. If active compression of the distended lung by the employment of expiratory muscles produces a vicious cycle of expiratory stenosis when there is bronchiolar hypertonus, then there is no reason apparent why the same process should not operate when the resistance to expiration is located in the trachea.

According to the prevailing theory, in the presence of great resistance to expiration located in the trachea there should be pulmonary emphysema and a prolongation of expiration proportionately greater than the prolongation of inspiration, but the clinician rarely has an opportunity to investigate this theory. By the time the patient with tracheal stenosis comes under observation there is nearly always a complicating tracheobronchitis with infection of the bronchial tree. What the clinician usually sees is tracheobronchitis complicating tracheal stenosis. Under these conditions the patient may have much moisture in the air spaces and there may be a complicating bronchiolar hypertonus, so that it is quite impossible to separate the effects of the two lesions so far as the clinical manifestations are concerned. I have seen patients with laryngeal diphtheria who had pulmonary emphysema, but these same patients

had tracheobronchitis as well as the laryngeal stenosis, so that one would not be justified in saying the patient's emphysema was the result of laryngeal stenosis. It could just as well originate from the bronchitis. However, during the influenza epidemic of 1919 I had an opportunity to see a case of uncomplicated tracheal stenosis:

A well-grown girl, about seventeen years of age, had been ill for three days with epidemic influenza. There had been a very considerable amount of tracheitis from the start. There was some dulness at the base of the right lung, but there was no elevation in the pitch of the respiratory sounds, and the patient was not cyanotic. Until the evening of the third day no alarming symptoms had developed, but about 10 o'clock on the night of the third day of her illness she became quite dyspneic. I saw her at 12 o'clock at night. She then had severe stenosis of the trachea, about halfway between the glottis and the bifurcation, which was due to edema of the tracheal mucosa. With the laryngoscope the livid swollen mucosa and the very narrow slit in the lumen of the trachea could be very plainly seen. This patient was doing her utmost to breathe. She employed a violent effort of all the inspiratory muscles during inspiration, and likewise employed violent contraction of all her abdominal muscles and intercostals during expiration. There was no undue prolongation of the expiratory phase and there was no evidence of increase in the volume of the lung. The patient died about two hours later from respiratory exhaustion due to stenosis of the trachea, but there was not the slightest evidence of emphysema.

Now it might be objected that in this particular patient the equality of resistance to inspiration and expiration was the reason why she did not have emphysema, but later we had the opportunity of seeing a patient who had an expiratory resistance in the trachea which greatly exceeded the resistance to inspiration, and there was no pulmonary emphysema:

A man having an aneurysm of the ascending arch of the aorta, which compressed the trachea at the bifurcation, was brought into the hospital in one of his attacks. It was quite apparent that the patient accomplished his inspiratory act with comparatively little effort. There was some resistance to inspiration, but the inspiratory dyspnea was not excessive, and it was quite plainly apparent that there was less dyspnea during the latter part of inspiration than at the beginning of the phase. In other words, as the man lifted his thoracic cage the compression of the trachea

lessened, but during expiration it was equally apparent that the resistance to the exit of air increased as the expiratory phase progressed. The expiratory phase was greatly prolonged and was attended with violent contractions of the abdominal and intercostal muscles. In fact the man was employing his utmost effort to expel air from his lung during the latter part of the expiratory phase. This was due to the fact that as the expiratory phase progressed and the thoracic cage was driven downward upon his aneurysm the stenosis of the trachea increased, so that the stronger his expiratory effort the more ineffectual it became. This was an instance of expiratory dyspnea, of a high degree, located in the trachea, but the volume of the patient's lung was not increased during the attacks. We had an opportunity to see him in several of these experiences, which were very distressing and seemed to threaten the patient's life, but the volume of the lung during the paroxysms (which lasted several hours) did not increase. This patient's expiratory dyspnea was just about as severe as a patient could endure and yet survive.

I could not conceive of a more successfully designed experiment to prove the fallacy of Biermer's theory of expiratory stenosis than this patient exhibited in his several paroxysms. It was this experience that suggested animal experiments to see what effect obstruction to inspiration and expiration, respectively, may have on the volume of an animal's lung when the seat of the obstruction is in the trachea.

Experimental Tracheal Stenosis. A wooden box, 31 x 10 x 9 inches, was constructed with a shelving top, which enabled us to seal the open top of the box with a glass plate laid in vaseline. With an animal confined in this box, which served as a plethysmograph, we could connect the cannula inserted in the animal's trachea with the exterior through a tube, which was passed through a rubber cork placed in a hole in the side of the box. Through a Meltzer cannula inserted in the pleural cavity we were able to register the intrapleural pressure by means of a tambour, which was connected with the tube leading from the Meltzer cannula through a cork in the side of the box. By the same means we connected a cannula in the dog's jugular vein with the exterior. The animal could breathe the room air or any other atmosphere, and by connecting the cavity of the box with the spirometer of a Benedict apparatus we could record the respiratory excursions and detect any modifications in the volume flow of air during inspiration and expiration

quite as accurately as when the tracheal cannula was connected with the spirometer. In fact the plethysmographic excursions of the spirometer were perfect reproductions of the excursions of the spirometer when the animal breathed directly into the reservoir. Each millimeter of elevation of the reservoir represented a volume of 22 cc. When the spirometer was used for recording the plethysmographic excursions the tubes leading to the canister were cut off, so that there was simply a to-and-fro movement of air between the Benedict reservoir and the air of the box which contained the dog. We could thus accurately measure not only the character of the inspiratory and expiratory excursion and its exact volume, but we could accurately measure any variation in the minimum volume of the animal's lung.

The tracheal cannula connected through the side of the box with room air, and on the end of this tube one could fix various sorts of appliance. For instance a screw clamp was used on a piece of rubber tubing which was attached to the tube connected with the tracheal cannula, and in this way we could give the animal any degree of stenosis to inspiration and expiration alike. By a Y-tube fixed to the tracheal cannula and clapper valves we could give the animal any degree of stenosis to expiration only and leave inspiration unobstructed, or we could do the reverse and give the animal any degree of resistance to inspiration and leave expiration unobstructed.

The animals employed varied greatly in size, the largest weighing seventeen kilos. Many times there was no change in the volume of the animal, but we never got an increase in volume above 66 cc when tracheal stenosis to inspiration or expiration was employed.

The dog was given chloretone (dissolved in oil) in the peritoneal cavity and morphin was given hypodermically. We attempted to use the minimum dose of chloretone and morphin, but it was necessary to keep the animal at rest while fixed within the plethysmograph.

One of the greatest difficulties was to procure active expiration in an anesthetized animal. Obstructing respiration and permitting the carbon dioxide to accumulate within the body, or having the animal rebreathe into a bag containing 80 per cent O_2 and 20 per cent CO_2 would not secure active expiration. Resistance to inspira-

tion very readily activated the muscles to increased effort, but when tracings of intrapleural pressure were made we found that resistance to both inspiration and expiration was not sufficient to induce active expiratory effort. *It was only after a high degree of hyperpnea was induced by having the dog rebreathe an atmosphere of oxygen and carbon dioxide that we could induce an active expiratory effort by shunting in a resistance to expiration.* This is mentioned because in the literature on the subject of respiratory excursions many writers have assumed that because respiration had been suspended or the animal was compelled to breathe an atmosphere with a high concentration of carbon dioxide, therefore the hyperpnea was accompanied by an active expiratory effort. *It was found after many trials that to procure an active expiratory effort, so that during expiration the pressure in the pleural cavity would be maintained above barometric pressure, it was necessary first to induce a very active hyperpnea and then during the period of hyperpnea to shunt in the resistance.* Under these circumstances the threshold for active respiration was passed and a positive pressure in the pleural cavity was obtained above that of barometric pressure.

Experiment. Fig. 1, line *A*, should be read from right to left. From 1 to 3 we see four normal respiratory excursions recorded by the plethysmograph. Line *A* is the base line and marks the minimum volume of the dog at the end of expiration. At 2 the animal was made to rebreathe into a bag containing 80 per cent O_2 and 20 per cent CO_2. Each 5 mm. in the time marker represents one second, so that 1 mm. equals one-fifth second. As can be seen by the tracing, inspiration and expiration were of about the same duration; and while the animal was breathing room air one second was occupied to complete the respiratory cycle, and the animal breathed about 70 cc with each respiration.

When the dog began rebreathing the high concentration of CO_2 the time occupied in the respiratory cycle was one second; the rate was very much increased and the volume of each excursion was 286 cc. From 2 to 4 the animal was rebreathing the CO_2 without resistance to either inspiration or expiration, and then at 4 a valve was inserted in the end of the tube connecting with the tracheal cannula, which gave the animal a perfectly free inspiratory movement but very greatly restricted the expiration. The volume flow through the expiratory valve under a pressure of 80 mm. of water was 1200 cc per minute. Inspiration occupied three-fifths of a second, but the duration of the respiratory cycle was thirteenth-fifths of a

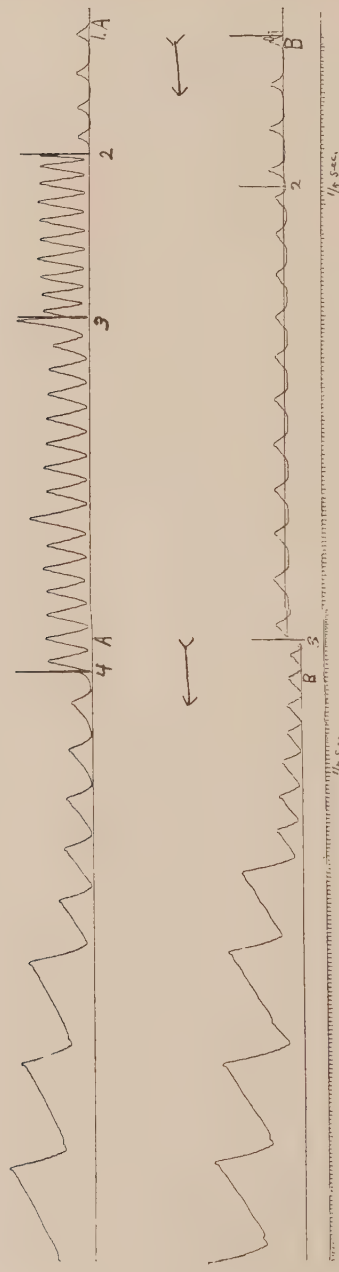

FIG. 1

second, so that the proportional duration of inspiration to expiration was 3 to 10. The volume of excursion as the animal began breathing at 4 was 132 cc. As will be seen by the tracing, in the first five respiratory cycles there was a gradual rise in the volume of each respiration, and then it is seen that the stimulus to respiration due to the CO_2 accumulation produced a large excursion of 400 cc. The duration of inspiration in this cycle was five-fifths of a second, but the duration of expiration was twenty-five-fifths of a second, and the volume of the dog at the end of expiration had increased 154 cc. As this tracing was continued the respiratory excursion became more rapid and the minimum volume of the dog at the end of expiration had increased about 250 cc above the original volume. So that we can say that this experiment shows that when an expiratory obstruction is shunted into the trachea in an animal with great hyperpnea the residual air in the lung is very greatly increased. When, however, one carried out the experiment in reverse order, and as shown in line B the dog breathed room air from 1 to 3 and then at 2, the expiratory valve was placed in the end of the tracheal cannula, so long as the dog was breathing room air and there was no occasion for hyperpnea the volume of the dog was unchanged. At 3, however, the animal continued to breathe with an expiratory resistance when he was connected with a bag containing 80 per cent O_2 and 20 per cent CO_2. By the end of the seventh respiratory cycle there was a large inspiration of 352 cc, and the same increase in the residual air of the dog became apparent in line *A* at 4. As seen in one of the larger excursions the duration of the respiratory cycle was thirty-fifths of a second, inspiration occupying only four-fifths of a second and expiration twenty-six-fifths of a second.

Although in this experiment the intrapleural pressure was not measured (because I wished to leave the thorax intact) I think we are justified in assuming from repeated trials in former experiments that with this resistance to expiration added to the great hyperpnea from breathing CO_2 the animal was actually employing effort during the expiratory phase. This experiment shows that the only way in which tracheal resistance will increase the residual air in the lung is in conjunction with increase in the volume of respiratory excursion. In this animal the volume of excursion was increased from 70 cc up to 300 cc by rebreathing CO_2 and the rate of respiration was increased from 36 to 52 per minute.

The phenomena described in this experiment do not occur in bronchiolar spasm. A patient who has a violent spasm can supply his oxygen needs perfectly well by a 25 per cent increase in the

oxygen consumption and in the minute volume of air. I have repeatedly taken Benedict tracings on patients during periods of bronchiolar hypertonus, and during severe spasm we have found both the minute volume of respired air and the consumption of oxygen just about 25 per cent greater than in the interims of respiratory comfort. The increase in each is imposed on the patient by the additional work required to ventilate his lungs. This introduces another essential consideration of asthmatic breathing—that is, that the residual air in the lung is greatly increased, although the increase in the minute volume of ventilating air is slight. In bronchiolar hypertonus without hyperpnea emphysema is produced, but in tracheal expiratory dyspnea emphysema is obtained only in the presence of hyperpnea, which must be superadded to the expiratory resistance.

The clinical and experimental evidences reveal an essential difference between the results of bronchiolar hypertonus and stenosis of the trachea. When the tracheal resistance to inspiration and expiration are equal there is no increase of residual air in the lung. In the absence of hyperpnea, resistance to expiration in excess of that to inspiration will not increase the volume of the lung.

Bronchial Hypertonus, Regional and of any Degree. The Biermer doctrine of the mechanism of emphysema in bronchiolar hypertonus demands an active expiratory compression of the lungs. But moderate bronchiolar hypertonus produces emphysema when there is no active effort attending expiration. Expiration may remain a passive procedure and still be attended with emphysema. When the conception of emphysema from bronchiolar hypertonus first appealed to me as a problem for investigation, I was in the habit of thinking of bronchiolar hypertonus as a neuromuscular performance that belonged to the all-or-nothing law and also that it was equally distributed throughout the bronchial tree. Neither of these conceptions is true. Neuromuscular reaction in the bronchial tree may, like vasomotor reaction in the arterial bed, be of any degree and may also be universal or regional. A patient may have a moderate bronchiolar hypertonus attended with demonstrable emphysema of the lung but without sufficient bronchiolar resistance to call out an active expiratory effort on the

part of the patient. A few clinical cases will suffice to prove these statements:

Case 1.—A man, aged sixty years, who had suffered from bronchitis and occasional asthmatic paroxysms for a period of five years, had a very moderate pulmonary emphysema, which at the borders of the lungs was sufficient to fill the pleural sinuses; but the total lung volume was not increased, so that the diaphragm was sufficiently flattened to change the normal direction in the movement of the costal margins during inspiration. According to this man's account his breathing was never completely shunted out of the field of consciousness. He was always aware of a certain degree of respiratory discomfort, but from physical examination and *x*-ray pictures of the lung one could not determine how much of his discomfort should be ascribed to atrophic emphysema and how much to bronchiolar hypertonus. When he was given a subcutaneous dose of 20 minims of 1 to 1000 adrenalin the vital capacity of his lung increased from 2000 to 2500 cc, and he then said that for the first time in a year he experienced complete respiratory euphoria. Before the adrenalin was given this patient did not have an active expiratory phase, although he had a certain degree of bronchiolar hypertonus with consequent emphysema, as shown by the respiratory comfort and increase in vital capacity directly after adrenalin was given. His respiratory discomfort had evidently consisted in an increased effort during inspiration. Of this effort he was conscious, but his pulmonary ventilation was still adequate to preserve the passive character of his expiration. His expiration was prolonged, but the moderate dyspnea permitted the expiratory prolongation.

Case 2.—Another patient was a young, vigorous man, aged twenty-five years, who complained of respiratory discomfort which had lasted for twenty-four hours. It was quite apparent that the man had labored breathing, but examination revealed that his labor was all on the inspiratory side; there was no effort during the expiratory phase, the abdominal muscles being perfectly relaxed. The costal margins moved outward throughout their extent during inspiration. There was sufficient emphysema, however, at the borders of the lungs, so that they filled the pleural sinuses as far as the eighth interspace in the nipple line and the eleventh rib in the axillary line.

This patient showed marked relief from the administration of adrenalin. Before the administration his vital capacity was 3350 cc when sitting and 2951 cc when lying. Ten minutes after he had been given 20 minims of 1 to 1000 adrenalin hypodermically his vital capacity when sitting was 3478 cc and 3415 cc when lying. His respiratory rate both before and after adrenalin varied between 22 and 24. Before he had his adrenalin,

when the respiration was traced on a rotating drum which was running at the rate of 5 mm. per second, in a respiratory cycle during which he breathed 528 cc, the duration of the entire cycle was twenty-two-fifths of a second. The inspiratory phase was ten-fifths of a second and the expiratory phase twelve-fifths of a second. After he had adrenalin, in a respiratory cycle in which he breathed 726 cc, the duration of the cycle was nineteen-fifths of a second, the inspiratory phase occupying eight-fifths of a second and the expiratory eleven-fifths. So it is quite apparent that both before and after adrenalin there was no active expiratory phase. Although the test showed only a comparatively slight increase in his vital capacity after adrenalin the lung passively expelled 726 cc in eleven-fifths of a second, whereas prior to the administration of adrenalin the lungs passively expelled 528 cc in twelve-fifths of a second. The rigidity of the lung was considerably less after adrenalin than before. The inspiratory effort had been quite apparent to inspection and the inspiratory and expiratory phases were accompanied by a number of coarse, moist and sibilant rales, all of which disappeared after adrenalin took effect. The only direct evidence we procured to show diminution in volume of the lung after adrenalin was that while the lower border of the lung had been at the eleventh rib in the axillary line before adrenalin it was afterward at the ninth interspace. In the midclavicular line it was at the eighth interspace both before and after adrenalin was given. However it was quite apparent to the observer that the adrenalin relieved the man of a very considerable degree of respiratory discomfort.

These patients with demonstrable bronchiolar hypertonus had a very considerable degree of respiratory discomfort (that is, inspiratory), but the vital capacities of the lungs were very little modified by the hypertonus. According to the size and conformation of the second man's thorax he should have had a vital capacity of at least 5000 cc. Without the use of adrenalin it would have been impossible to show that bronchiolar hypertonus played a part in his respiratory discomfort.

The following cases showed regional hypertonus:

Case 3.—This patient had had a moderate general bronchiolar hypertonus, but on one occasion it was only regional. The resident physician found one night upon examination that his right thorax was comparatively immobilized, with no respiratory excursion perceptible in the arches of the right ribs. The whole right thorax was distinctly larger than the left. There was a distinct asymmetry which had not been visible on other occasions. During the inspiratory phase the entire right costal border was

drawn strongly toward the median line, but the left moved vigorously in a lateral direction and the respiratory excursion of the entire left thorax plainly exhibited an exaggerated excursion. The arches of the left ribs moved in their normal bucket-handle manner much more actively than during interims of comfort. It was plainly apparent that the right thorax was enlarged, that the right diaphragm was flattened and that there was very slight respiratory excursion on that side. It was also apparent that there was a compensatory increase in the respiratory excursion of the left side, the minute volume of air in the left lung being equal to that of both lungs in the interims between attacks. In other words the man had an acute unilateral bronchiolar spasm. This was also confirmed by auscultation. Over the entire right side there was an abundance of squeaking and moist rales during inspiration and expiration, but the left lung was free. The respiratory sounds were only faintly audible over the right side, whereas they were distinctly heard over the left, where the excursion was magnified. This patient was probably breathing a little larger minute volume of air than he employed during his interims of comfort, but the greater part of the pulmonary ventilation was accomplished with the left lung.

Twenty minims of 1 to 1000 adrenalin were given this patient hypodermically, and within a very few minutes he recovered his normal thoracic excursion. The two sides were again symmetric, with symmetric outward movement of both costal margins during inspiration. Most of the rales of the right side disappeared. The patient was perfectly comfortable, and so far as physical examination enabled us to determine he was again ventilating both lungs equally.

Case 4.—This patient had severe bronchitis and atrophic emphysema, and he had been studied during very many asthmatic paroxysms. On one occasion, however, he complained of discomfort when the auscultatory evidence of asthma was limited to the lower lobe of the right lung. The right costal border was drawn toward the median line during inspiration, whereas the left costal margin moved in an outward direction. After the hypodermic injection of adrenalin the patient's respiratory comfort was restored within a very few minutes. The auscultatory signs of asthma disappeared from the lower lobe of the right lung and the right costal border resumed its outward inspiratory excursion, which was quite symmetric with that of the left side. So far as physical examination enabled us to determine this patient had a regional bronchiolar spasm attended with emphysema restricted to the lower lobe of the right lung.

Case 5.—On another occasion we had a patient suffering from chronic bronchitis and emphysema who had an attack of asthma in which all the evidence of asthma was limited to the two upper lobes. There were no evidences of flattening of the diaphragm, but the auscultatory signs of

asthma, though very pronounced in the upper lobes, were entirely wanting over the two lower lobes.

It does not seem possible that a patient could produce the expiratory vicious cycle which supposedly occurs in asthma and confine the exhibition of compression of the bronchioles in one instance (Case 3) to the entire right lung, in another instance (Case 5) to the two upper lobes, and still in a third instance (Case 4) to the lower right lobe only. Such cases alone offer sufficient evidence to disprove the need of an active expiratory phase to produce emphysema during an asthmatic attack.

Duration of Expiration. To return to our cases of general bronchiolar hypertonus (Cases 1 and 2) attended with moderate emphysema and prolongation of the inspiratory phase. The expiratory phase in all the patients with moderate hypertonus was longer than the inspiratory phase simply because the resistance to expiration was not sufficient to demand an active compression of the lungs. To cause a transition from passive to active expiration the obstruction to the passage of air must be so great that the time the passive expiration requires exceeds the tolerance of the patient's respiratory needs. So long as bronchiolar hypertonus will allow a passive expiratory phase the patient will instinctively refrain from active compression of the lungs by simultaneous action of the abdominal and intercostal muscles. The active expiration demanded by stenosis of the bronchial tree is very exhausting. Resistance to inspiration requires only an increase in the force of activation of the muscles normally employed, but resistance to expiration not only requisitions muscles which are not normally employed (which in itself is very exhausting) but produces an unfavorable effect on the hydraulics of the blood circulation in the thorax. In animal experiments the extremity of the measures which are required —namely, hyperpnea plus obstruction—before the animal can be induced to raise the expiratory pressure above the barometric pressure plainly illustrates the height of this threshold for active expiration. These facts clearly explain why a patient with moderate bronchiolar hypertonus will have the duration of the expiratory phase shortened after the administration of adrenalin. Adrenalin given to such a patient will also have the effect of diminishing the

residual air in the lung and will increase the pulmonary vital capacity.

How is the expiratory phase affected in a patient who has a bronchiolar hypertonus so severe that an active expiration is demanded? In Fig. 2 is a tracing taken with the Benedict apparatus during an attack of bronchiolar hypertonus. On line *B* is a tracing after the hypertonus was relieved by 20 minims of 1 to 1000 adrenalin given

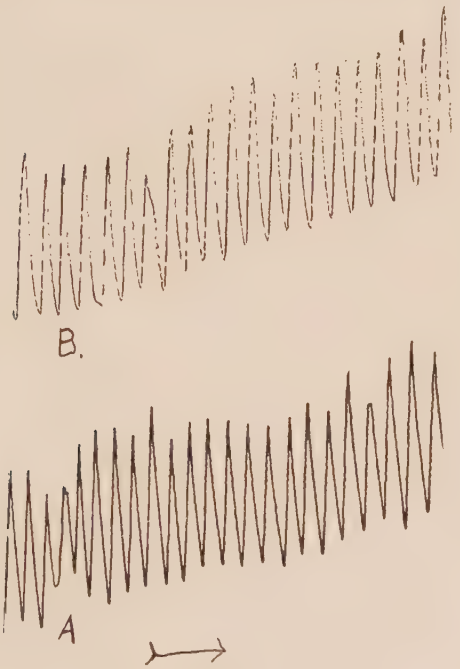

Fig. 2

hypodermically. When the tracing in line A was made the patient was in great distress. The volume of each lung was so increased that the flattened diaphragm drew the costal borders toward the median line during inspiration and the expiratory phase was accomplished by violent contraction of the abdominal muscles. These conditions are exactly such (according to Biermer's doctrine) as to create a vicious cycle of dyspnea and should have caused a prolongation of the expiratory phase with a lessened volume flow of air

toward the latter part of expiration. But the tracing reveals the opposite of that expected under the Biermer theory.

Experiment. This tracing was taken on a drum with the speed of 1 mm. per second. The respiratory rate before adrenalin was given was 16 per minute. The line of the inspiratory phase on the upstroke is straight and the line of the expiratory phase is equally straight. There is no lagging or horizontal drag as must occur in such a tracing should the volume flow lessen toward the end of expiration. The transition from inspiration to expiration and that from expiration to inspiration show very sharp peaks. The tracing indicates just what we were able to observe in the patient at the time. He was employing his utmost muscular strength during all of inspiration and expiration, and the volume flow of air was uniform throughout the entire respiratory cycle.

On line *B* we see the effect on the respiratory cycle after the bronchiolar spasm had been relieved by the hypodermic injection of 20 minims of 1 to 1000 adrenalin. The respiratory rate was then 14 per minute. The inspiration on the upstroke is not quite so nearly vertical as before the adrenalin was given, and we can see toward the end of the expiratory phase a distinct slope to the right, which means that the volume flow toward the end of the expiratory phase was distinctly lessened. During the time the tracing in line *B* was taken the patient had distinct inspiratory widening of his subcostal angle. Expiration was not attended with contraction of the abdominal muscles. In fact he was breathing with perfect comfort. The difference was that before he had his adrenalin he was able to attain his required pulmonary ventilation at the expense of great muscular effort during both inspiration and expiration. The expiratory labor was very exhausting, although the actual increase in oxygen consumption was only 25 per cent in excess of what he employed during tranquil breathing at rest. The source of exhaustion is not in this instance measurable by the increased consumption of oxygen but by the employment of an unusual kind of respiratory effort. With the Biermer doctrine of expiratory dyspnea in mind one would be disposed to say that line *B* was traced before adrenalin was given and that line *A* was traced after the bronchiolar hypertonus had been relieved. This tracing shows a longer expiratory phase after bronchiolar hypertonus was relieved simply because the patient had a passive expiratory phase and his respiratory needs did not demand compression of the lungs during expiration. Tracing *A* not only shows that the patient employed a positive pressure in his pleural cavity during the expiratory phase, but it shows that the minute volume flow of air during the latter part of the expiratory phase was better maintained than when he was breathing with a passive expiration.

These experiments not only prove the fallacy of the doctrine of expiratory dyspnea in asthma, but they also show the error of employing the prolongation of the expiratory phase as an index of the severity of bronchiolar spasm.

Expiratory Compression Stenosis of the Bronchi. The question of expiratory dyspnea arising from compression stenosis in the bronchial tubes remains for experimental consideration. If bronchiolar spasm is severe enough to require active compression of the lungs in expiration there are two forces employed to expel air from the lungs: one the positive intrapleural pressure due to activation of the abdominal and intercostal muscles, and the other the retractile property of the visceral pleura and the elastic tissue within the lungs. If active compression of the lungs produces stenosis of the small branches of the bronchi in the presence of bronchiolar spasm, why should it not do the same in the absence of bronchiolar hypertonus? It seems reasonable to suppose that in the absence of hypertonus the bronchi would be more compressible than when they are hypertonic. If hyperpnea is induced in an animal by suffocation or by breathing a high concentration of CO_2, and then tracheal stenosis is added to the respiratory burden so that compression of the lung is required to accomplish expiration, then under such experimental conditions we can measure the volume flow of air from the trachea and also the pressure on the proximal side of the tracheal stenosis, and synchronously with these air pressure and air volume flow measurements we can also trace the expiratory pressure in the pleural cavity. In such an experiment the expiratory forces are pitted against the tracheal resistance, with a certain volume flow as a result. Should an expiratory resistance to the outflow of air intervene between the air cells and the tracheal obstruction, then the volume flow of air should lessen toward the latter part of expiration and the sum of the expulsive forces should exceed the air pressure maintained in the trachea on the proximal side of the stenosis. The expulsive forces are, of course, the pressure within the pleural cavity and the retractile power of the lung.

Experiment. A dog, weighing 16 kilos, was anesthetized with chloretone and morphin. Tracheotomy was then done and two cannulæ introduced,

one into the trachea and one into the right pleural cavity, after which the dog was placed in the plethysmograph. The cavity of the box was connected with the reservoir of a Benedict apparatus and the passages to the canister clamped so that there was a free movement to and fro between the confined air in the box which contained the dog and the air in the registering reservoir. The volume flow of air during each respiratory cycle could thus be accurately traced on a revolving drum, which had a speed of

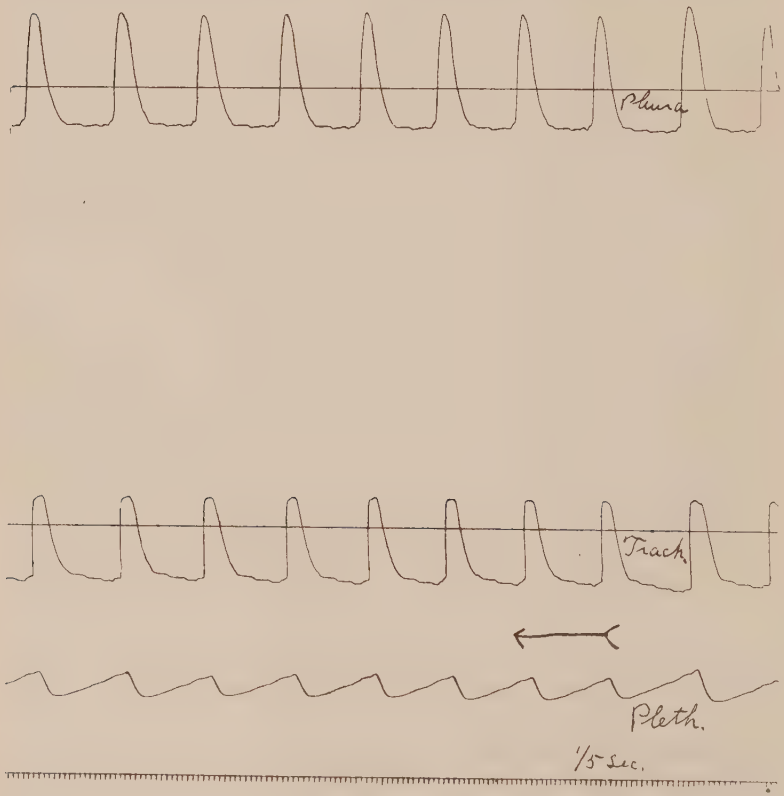

FIG. 3

5 mm. per second (Fig. 3, line *Pleth*). The time is marked in fifths of seconds. By a tube which passed through a cork in the side of the box the Meltzer cannula in the pleural cavity was connected with a calibrated tambour which traced the pressure in the pleural cavity for each respiratory cycle (see line *Pleura*). A T-tube was inserted in the tube to the pleural tambour, by means of which we could introduce into or remove from the pleural cavity any desired quantity of air. The tracheal cannula

was connected with a tube which passed through a cork in the side of the box, and to the end of this tube was fitted a Y-tube, the afferent limb of which was fitted with a clapper valve while the efferent limb had its lumen modified at will by means of a screw clamp.

On the proximal side of this Y-tube, which terminated the tracheal cannula, was introduced a T-tube, which was connected with a calibrated tambour by means of which could be traced the air pressure on the proximal side of the expiratory obstruction. The writing styles for the plethysmograph and tracheal and pleural tambours were then perfectly aligned, so that a vertical line drawn anywhere would enable us to make accurate comparisons of the three tracings in any phase of a respiratory cycle. In both the pleural and tracheal pressure tracings the horizontal line traces the line of barometric pressure. The time tracing is in fifths of seconds. In all three tracings the up stroke records inspiration and the down stroke expiration, and they are read from right to left. They were made after 1050 cc of air had been injected into the right pleural cavity. This injection was to procure any evidence of expiratory bronchiolar compression which the partial collapse of the lungs might offer.

Preceding this part of the tracing the screw clamp on the efferent limb of the tracheal Y-tube was screwed down to procure the maximum resistance against which the animal would breathe. Respiration was arrested by complete obstruction of the tracheal cannula for one minute and the drum was started as the complete obstruction of the tracheal cannula was released. The afferent clapper valve offered slight resistance to inspiration, as shown by the rise of the pressure curve of the tracheal tambour above the line of barometric pressure (minus pressure). The combination of hyperpnea with resistance in the efferent limb of the Y-tube to the tracheal cannula induced an active expiratory phase, as shown by the rise above barometric pressure in the pleural cavity (below the pleural barometric line).

The plethysmographic tracing shows each respiratory cycle to have a duration of seventeen-fifths of a second, the inspiratory phase occupying three-fifths of a second and the expiratory fourteen-fifths. The volume of each respiration was 110 cc. The volume flow tracing of expiration is straight; there is no drag to the left as there would be if the flow diminished toward the latter part of the expiratory phase.

When the positive pressure below the barometric lines of pressure for the trachea and pleura are measured the pleural pressure of 9 mm. registered by the tambour style is found to equal 70 mm. of water, and the 13 mm. registered by the tracheal tambour style at the same time equals 100 mm. of water. But to the air pressure within the pleural cavity must be added the elastic contractile force of the lung. In this dog the intrapleural pressure measurements were taken when the animal was breathing freely without obstruction and the pleural cavity was free of all but 50 cc of air. The

intrapleural tracings then showed a pressure below the barometric pressure of 70 mm. of water at the height of inspiration and 30 mm. of water at the end of expiration. If these 30 mm. are added to the maximum intrapleural pressure attained during the expiratory phase we find the sum of the two factors (70 + 30) exactly equals the positive pressure measured on the proximal side of the screw clamp on the efferent limb of the tracheal cannula, and furthermore the minute volume flow is constant during the entire expiration. *We are justified in saying that under these experimental conditions there can be no resistance to the exit of air intervening between the air cells and the point of obstruction on the efferent limb of the tracheal cannula.*

Repeatedly hyperpnea was produced by having the animal rebreathe into a bag containing 80 per cent O_2 and 20 per cent CO_2, and then resistance was applied in the tracheal path. By these means we were able to develop a high pressure in the pleural cavity during expiration and the same results were always procured. After each experiment the tambours were calibrated for pressures above and below barometric pressure. In the dog the mediastinal reflections of the pleura are so mobile and offer so little resistance that a positive pressure obtained on one side of the thorax by producing an active expiration will be the same on both sides.

In measuring the volume of an animal under these experimental conditions great care must be observed to guard against leakage of air into the pleural cavity. If air enters the volumetric measurements of the plethysmograph will record an increase in volume of the thorax, which apparently varies with the resiliency of the animal's thoracic cage. In one experiment 100 cc of air injected into the right pleural cavity increased the volume of the dog 120 cc, and when 500 cc were injected the dog increased 360 cc in volume. If this source of error is not considered the increase in volume may be interpreted as an increase of the residual air in the lungs.

Thus far the studies of tracheal obstruction show several results that are of interest in relation to the study of resistance to the respiratory passage of air:

1. If the obstruction is the same during inspiration and expiration there will be no increase of residual air, although hyperpnea is demanded for breathing an atmosphere of 80 per cent O_2 and 20 per cent CO_2.

2. When there is no hyperpnea equal resistance to inspiration and expiration fails to produce emphysema.

3. In an animal under chloretone anesthesia the threshold for active expiration is very high and is passed only when hyperpnea is added to tracheal resistance.

4. Expiratory resistance with unobstructed inspiration will not cause emphysema unless the resistance occurs in company with hyperpnea.

5. When active expiration is induced by the combination of hyperpnea with tracheal expiratory resistance there is no intervening resistance to the exit of air from the lungs that can be located in the bronchial tree, as Biermer's theory assumes there should be to produce emphysema.

The above experimental results are quite consistent with the previously described observations on a patient who suffered from severe stenosisof the trachea (see p. 68), whichcaused equal resistance to inspiration and expiration; and they are also consistent with the observations made on the patient who had severe expiratory with moderate inspiratory resistance without the development of emphysema (see p. 68), although the expiratory phase in both patients was accomplished with the utmost prolonged muscular effort of which the patients were capable.

Acute emphysema was procured by animal experiment only when the hyperpnea was combined with expiratory dyspnea in the absence of inspiratory dyspnea. On the other hand, we have shown that emphysema with bronchiolar hypertonus occurs in man when there is no further cause for hyperpnea than a slight effort during inspiration, and when expiratory dyspnea is so moderate that an active expiration is not required and expiration remains a passive procedure.

Experimental Bronchial Hypertonus from Histamine. We should then expect to find emphysema in dogs accompanying the bronchiolar hypertonus that follows intravenous injection of histamine. With this in mind, twelve experiments were performed which were designed to show modifications in the volume of the lung of a dog during the period of bronchiolar hypertonus. The histamine was administered through a cannula which was previously

fixed in the internal jugular vein, and a tube from the cannula led through a cork in the side of the box, so that we could give histamine without disturbing the plethysmographic tracings of respiration. I was never able to find an increase of lung volume above 60 cc during the period in which the effect of histamine was apparent, and that was in a dog weighing 16 kilos. This increase in size is practically negligible so far as a consideration of increase in the residual air of the lung is concerned, for such a moderate increase might occur under a variety of disturbances in the cardiorespiratory function.

Experiment. In Fig. 4 there is a tracing from an experiment which was designed to show the intrapleural pressure during the period of the histamine effect, with the idea that if histamine produced a decided bronchiolar hypertonus then there should be a lessened respiratory excursion in the presence of a lowering minus pressure within the pleural cavity. In this experiment the plethysmograph was not used. The animal breathed through a tracheal cannula directly into the Benedict reservoir. The tracing reads from left to right and the time-marker is in fifths of seconds. Lines A and A' were taken simultaneously. The horizontal line marks the line of barometric pressure in the pleural tambour. Line A' was a simultaneous tracing in the Benedict apparatus.

After the injection of 5 cc of 1 to 10,000 histamine into the jugular vein the respiratory rate became very rapid and the volume of excursion was very small. To the right it will be seen that during the period in which the pleural tracing showed a very great descent in pressure the volume of tidal air in the lung diminished. In fact, where the excursion of pleural pressure was very considerably extended, from the barometric line to a considerable distance below, the excursion of the lung was quite small. So we are certainly justified in saying that in the presence of a low barometric pressure in the pleural cavity, and also in great excursions of the pressure within the pleural cavity, there was a marked rigidity of the lung.

Lines B and B' are tracings taken from the same animal after a second injection of the same dose of histamine. The beginning of the histamine effect is apparent at H in the tracings both of pleural pressure and of lung excursion. The two experiments show essentially the same thing.

Thus far our investigation has shown that equal tracheal stenosis to inspiration and expiration in both men and dogs failed to produce emphysema. It has also been shown in both men and dogs that when tracheal stenosis to expiration is greatly in excess of that to

inspiration there is no emphysema. It has further been shown in dogs that when tracheal expiratory resistance is much greater than resistance to inspiration, emphysema can be induced only with the addition of hyperpnea, as follows, of course, when the animal is made to rebreathe into a bag containing a high concentration of carbon dioxide.

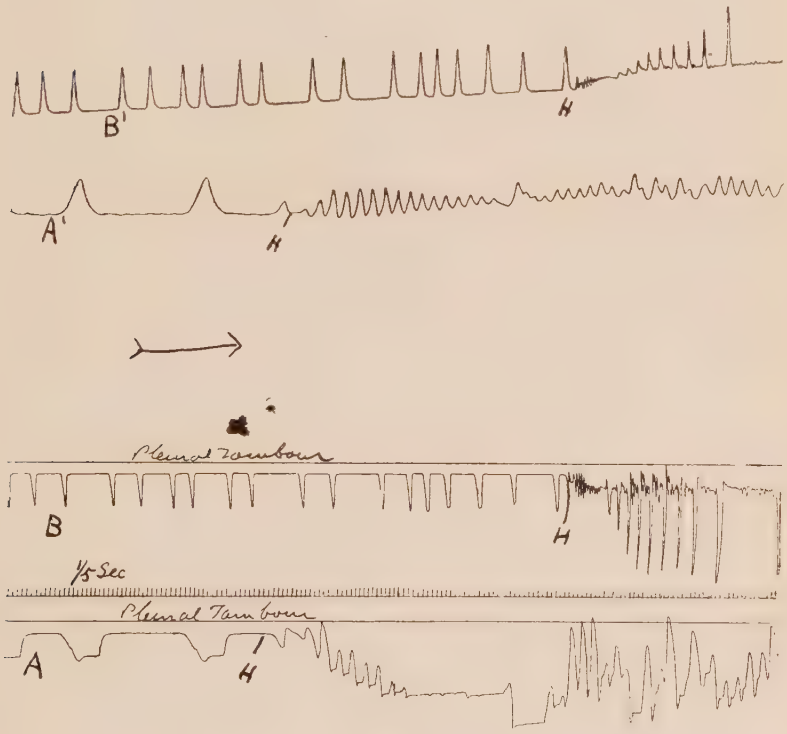

FIG. 4

Furthermore, the experiment in which the tracing of Fig. 4 was made proved that when a general bronchiolar hypertonus was produced by the use of histamine there was a great lowering of the barometric pressure in the pleural cavity during the entire respiratory cycle, but in spite of it there was a very small respiratory excursion and the total volume of the lung was not increased.

The following experiment was made for the purpose of tracing

the character of volume of the respiratory excursion and measuring the volume of the animal under the effect of a large dose of histamine with an intact thorax. The only operative procedure was the introduction of cannulæ into the trachea and the jugular vein, into the latter of which the histamine was injected:

Experiment. The tracing (Fig. 5) should be read from right to left and is simply the plethysmographic respiratory tracing of a 9-kilo dog. The first part, from 1 to 2, shows the animal breathing atmospheric air and then at 2′ the animal was made to rebreathe into a bag containing 80 per cent O_2 and 20 per cent CO_2. Under the influence of hyperpnea thus induced, with a perfectly free tracheal cannula, the total volume of the dog was increased about 50 cc. Then during this period of hyperpnea 10 cc of 1 to 10,000 histamine were injected into the jugular vein and immediately there followed a great modification in the respiratory excursion. Before the effect of the histamine was apparent and while the animal was rebreathing a high concentration of CO_2 the volume of respirations varied between 500 cc and 264 cc. Then before the histamine took effect the duration of the respiratory cycle was six-fifths of a second. After the histamine took effect the volume of the respiration was 110 cc and the time of a respiratory cycle was thirteen-fifths of a second, the expiratory and inspiratory phases having exactly the same duration. It is quite apparent from the prolongation of both inspiration and expiration, with a great diminution in the volume of the tidal air, that the animal was breathing against a great resistance in both inspiration and expiration, and although in this experiment there is no tracing of the intrapleural pressure, I believe we are justified in assuming from our former experiments that the animal was employing a vigorous respiratory effort. *The tracing shows that the volume of the dog was not increased, although the bronchiolar spasm was induced at a time when great hyperpnea was demanded by rebreathing 20 per cent CO_2; and just as we have seen in our patients with severe bronchiolar spasm when active expiration was demanded, inspiration and expiration are of equal length.*

This experiment, in the light of all the preceding observations, offers very conclusive evidence that a uniform hypertonus throughout the bronchial tree will not prolong the expiratory more than the inspiratory phase, and will not produce emphysema. Furthermore, in the experiment from which the tracing in Fig. 3 was made there is very satisfactory evidence that active compression of the lung does not create a vicious expiratory cycle by compressing the outlet of the respiratory units, as the Biermer theory demands.

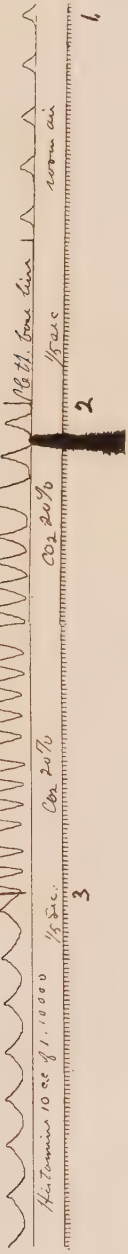

FIG. 5

The following case offers clinical confirmation of the foregoing statements:

The tracing shown in Fig. 6 was made from a young, vigorous man who had an attack of bronchiolar spasm which was not preceded by bronchitis, and so far as we are able to determine the bronchiolar hypertonus was a symptom of anaphylaxis from hay fever and not a hypertonus of the bronchial musculature which could be traceable to any nervous excitation within the bronchial tree or within the mediastinum. There was a con-

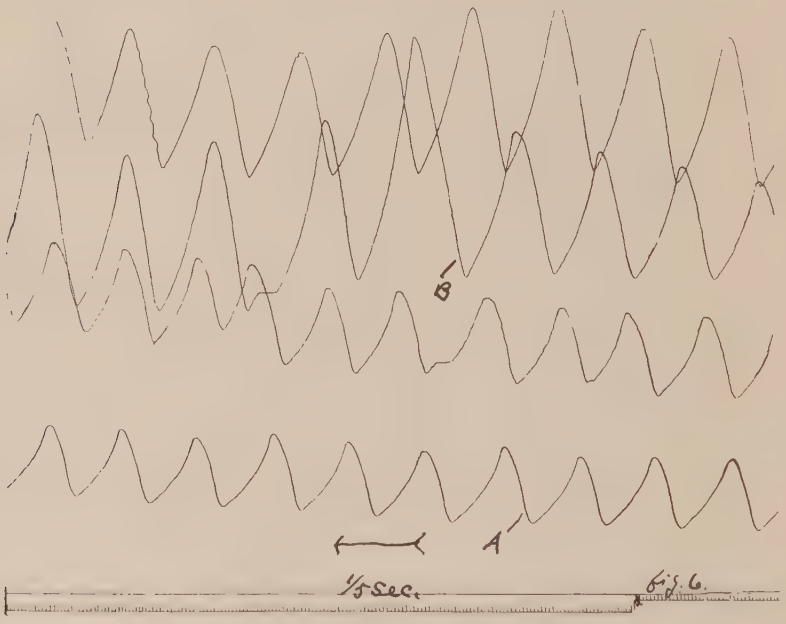

FIG. 6

siderable amount of nasopharyngeal irritation. The results of the tracing, which was made during his period of bronchiolar spasm and continued after adrenalin was given hypodermically until he had perfect respiratory relief, present very good evidence that in clinical experience we may have bronchiolar hypertonus which will greatly reduce the vital capacity of the lung and not increase its residual air. In fact we have in this patient with a uniform bronchiolar hypertonus quite the same results, so far as the vital capacity and lung volume are concerned, as have been shown in dogs when bronchiolar hypertonus was induced by intravenous injection of histamine.

This patient went to bed with perfect respiratory comfort and was

awakened in the middle of the night by a feeling of great respiratory discomfort. Inasmuch as the patient was one of the interns on the medical service in Lakeside Hospital he was very conversant with the methods of studying respiratory phenomena and was able to give ideal coöperation. The tracing should be read from right to left. The time-marker is in fifths of seconds. The two lower lines were traced before the effect of adrenalin was apparent and the two upper lines were traced after the bronchiolar hypertonus had been relieved by hypodermic injection of 1 to 1000 adrenalin.

Before adrenalin was given the vital capacity of his lung had been 2699 cc sitting and 1932 cc lying. After the effect of adrenalin was produced the vital capacity was 4264 cc sitting and 3893 lying. The respiratory rate before adrenalin was 18 per minute and afterward 14. Before adrenalin there was a great abundance of fine and coarse sibilant rales over each portion of both lungs, but after adrenalin all traces of adventitious sounds disappeared. His sense of discomfort was purely on the inspiratory side.

Expiration was a passive procedure, involving no contraction of the abdominal muscles. The patient said that on former occasions he had had attacks much more severe and exhausting, and during them he had been compelled to employ the abdominal muscles during expiration.

In the lower line at *A* the duration of the respiratory cycle is eighteen-fifths of a second. The duration of inspiration is six-fifths of a second, and, as will be noted in the tracing of the inspiratory phase, the up stroke shows a drag toward the left after the top of the excursion is approached. In other words there is evidence of a slowing of the volume flow toward the end of the inspiratory excursion. The expiratory phase lasted twelve-fifths of a second, just double the duration of the inspiratory phase, and the amount of air expelled during the twelve seconds was 374 cc. At *B*, after the bronchiolar hypertonus had subsided, we find a respiratory cycle which lasted twenty-four-fifths of a second, the duration of inspiration being eleven-fifths of a second and that of expiration thirteen-fifths, and the amount of air expelled was 2210 cc. So before adrenalin was given 324 cc were passively expelled in twelve-fifths of a second and after adrenalin was given 2210 cc were passively expelled in thirteen-fifths of a second. In spite of the fact that the vital capacity during the attack of bronchial spasm was lowered over 1500 cc, there was evidence of only a very slight increase in the volume of the lung. Before adrenalin the lower border of the lung was in the ninth interspace in the axillary line and after adrenalin it was in the eighth interspace; and during the attack, although his diaphragm was coördinately activated with his intercostal muscles, all of both costal margins moved in an outward direction during inspiration. So we are justified in saying that the reduction in vital capacity was not traceable to an increase in residual air in the lung but was due purely to bronchial stenosis.

Dr. L., from whom the tracing was taken, has repeatedly tested his vital capacity during the past year and has always found it to be about 4000 cc. In fact the character of this tracing shows his respiratory excursion during the attack to be quite like the respiratory excursions found in dogs when an equal tracheal stenosis to inspiration and expiration is employed. After the attack was over there was not the slightest evidence of any kind for bronchitis or any modification in the extensibility of the lung.

Regional Expiratory Dyspnea. At a stage of this investigation when the relation between bronchiolar hypertonus and pulmonary emphysema seemed very elusive a patient came into Lakeside Hospital who presented a group of symptoms that I had never before seen. This man had an aneurysm of the aorta involving the latter portion of the ascending and the first portion of the transverse arch. The first part of his aorta showed no evidence of dilatation. All the physical signs and the x-ray pictures confirmed the diagnosis of an aneurysm which lay in front and above the left bronchus and showed no evidences of contact with the right bronchus. In all cases of obstruction of the bronchus from aneurysm which had come under my observation there was an equal obstruction to inspiration and expiration, and there never had been in former cases any difference in degree of obstruction when the patient was sitting or lying. This man, however, showed a very curious phenomenon: When he was in the upright position there was no demonstrable stenosis to the left bronchus, but when he lay on his back there were decided evidences of stenosis to this bronchus, and during expiration it was much greater than during inspiration. The man suffered no pain and no great discomfort when he was going about. His complaint was that when he lay down he had an uncomfortable, "stuffy" feeling over the left side of his chest.

Physical examination of this patient revealed very characteristic signs of aneurysm of the arch of the aorta, as previously stated. When he was in the upright position the respiratory excursions of the two sides of the thorax were nearly equal, but when he lay down the left side became distinctly larger than the right and the respiratory excursion of the entire left side was very much less than on the right. Palpation revealed comparative immobilization of the ribs of the left side during inspiration in

this position and over the left hypochondrium there was an absence of the piston thrust from the left leaf of the diaphragm, whereas the right side of the diaphragm had a perfectly normal and very active excursion. The costal margin of the left side moved very slightly in an outward direction during inspiration, and percussion revealed a decidedly lower position of the left lung in the axillary line.

When the patient was upright the respiratory sounds on the two sides of the thorax were equally loud, but when he lay down those on the left side were only faintly audible. There was also much less tactile fremitus on the left side.

When he was sitting his vital capacity was 3711 cc; when lying, 2545 cc—a difference of 1166 cc, which is a far greater disparity than one will find in any other condition, for few persons will on lying down show a lessening of more than 250 cc in their vital capacity.

The pulmonary emphysema of the left side when the patient lay down was due to the relation between the aneurysm and the left bronchus. In this position the weight of the aneurysm compressed the left bronchus, but during inspiration the lifting of the thoracic cage permitted a freer flow of air into the right lung than that allowed out of the left lung during expiration. He was able to compensate for the impairment in ventilation of his left lung by a very moderate increase in ventilation of the right, and therefore had no air hunger. When the man was upright there was during expiration no demonstrable sign of compression of the left bronchus.

On comparing this case with the one described on page 68, in which there was an expiratory stenosis of the trachea from an aneurysm of the aortic arch, it will be apparent why this latter patient acquired an emphysema of his left lung when he was lying down. One can readily see that the essential difference between the two cases lies in the fact that the patient last described had a very decided expiratory dyspnea involving the left lung but none involving the right, whereas in the former patient, although he suffered from severe expiratory dyspnea, both sides of the lung were equally involved because the expiratory compression was applied to the trachea. This suggested the idea that pulmonary emphysema may be traceable to an excess of expiratory obstruction over inspiratory obstruction, and that clinically we find a marked pulmonary emphysema attending the process because the expiratory dyspnea is unequally distributed throughout the bronchial tree, whereas under the influence of histamine our experiments failed to show any emphysema simply because the hypertonus was equally distributed.

The patient with an expiratory dyspnea confined to the left bronchus had a great increase in the residual air of his left lung, which reduced the total capacity of his lungs by 1166 cc. This was accomplished without air hunger or hyperpnea or an active expiration, which is the very essential criticism above presented that must be met by a satisfactory theory for emphysema caused by bronchiolar hypertonus. If we conceive of these gross relations of expiratory dyspnea between the two lungs being applied to the terminal bronchioles throughout the bronchial tree, we then have a theory of emphysema from clinical bronchiolar hypertonus which conforms to the facts of clinical experience.

The physical signs produced by an aneurysm of the transverse arch compressing the left bronchus were very readily reproduced experimentally in the dog by procuring expiratory dyspnea confined to one lung with no obstruction to either bronchus during inspiration.

Experiment. A rubber cork was used, through the middle of which was passed a metallic tube with a diameter three-fourths that of the cork itself. This cork was just large enough to fill a bronchus of the dog employed. Over one end of the cork was spanned a small strip of rubber dam and the cork was so inserted into the right bronchus that this end lay in the distal position. During inspiration there was no obstruction to the entrance of air into the right lung, but during expiration there was very pronounced obstruction. Under a pressure equal to 80 mm. of water the minute volume flow of air through this valve measured 1200 cc, which would just about balance the elastic retraction of the dog's lung.

Fig. 7 shows an *x*-ray plate of the animal's thorax when he was comfortably breathing atmospheric air. The disparity in the elevation of the two leaves of the diaphragm was quite like that in our patient with aneurysm when he occupied the horizontal position. When the animal was made to rebreathe a high concentration of carbon dioxide this disparity was very considerably increased.

If we consider the factors attending pulmonary emphysema as a result of bronchiolar hypertonus we shall see that the following must be considered: (1) There is always dyspnea, but it is not necessarily so pronounced as to cause great discomfort. (2) The process is not accompanied by hyperpnea. Even in the very

severe cases the minute volume of air is not increased more than 25 per cent. (3) An active expiratory phase is not essential for the production of emphysema.

An unequally distributed expiratory dyspnea would satisfy all these criticisms, as shown in our patient with expiratory dyspnea restricted to the left side, and the dog with expiratory dyspnea restricted to the right side. From our experimental and clinical observations, therefore, it seems reasonable to suppose that in the

FIG. 7

acute cases an unequal distribution of the expiratory dyspnea is essential for the production of emphysema. In the chronic atrophic cases also it is known to be unequally distributed.

This study does not pretend to offer a satisfactory solution of the problem of pulmonary emphysema. As in massive collapse of the lung, which is also dependent upon some effect on lung motility, our present knowledge of physiology is not sufficient for a satisfactory explanation. This study does, however, reveal the inadequacy of the commonly accepted theory of emphysema and justifies

the suspicion that an unequal distribution of expiratory stenosis throughout the bronchial tree is the essential factor.

Summary. 1. Though active expiratory compression of the lung is rarely employed the vigor with which the expiratory muscles can compress the lungs is greater than that with which the inspiratory muscles can distend them. Therefore the air inspired within a given time can be expired within the same time, provided the resistance in the trachea or the branches of the bronchial tree is the same in inspiration as in expiration.

2. When the tracheal or uniform bronchial resistance to expiration exceeds that to inspiration the residual air in the lung is increased only when hyperpnea attains such a degree that the respiratory need will not allow adequate time for the volume of the expired air to equal that of the inspired air.

3. Compression of the lungs in expiration does not produce a vicious cycle of increasing resistance to expiration.

4. Neither hyperpnea nor active expiration is essential for the production of emphysema.

5. Prolongation of expiration in emphysema does not measure the degree of expiratory resistance but indicates the patient's respiratory tolerance of prolongation of the expiratory phase. It is only in the extremity of respiratory needs that active expiration is employed to overcome expiratory resistance.

6. In bronchiolar spasm severe enough to demand an active expiration the inspiratory and expiratory phases have the same duration, and the volume flow within each phase is constant.

7. That an excess of expiratory over inspiratory resistance should produce emphysema the excess must be unequally distributed in the bronchial tree.

DISCUSSION

Dr. H. Sewall, Denver, Col.: Dr. Hoover spoke of displacement of the diaphragm. In our clinical observations we take little account of possible variations in the tonus of the diaphragm and its pathologic significance, as we have learned to do in the case of the heart muscle.

In a very septic woman, recently observed, the absolute liver dulness reached the fourth rib on the right side in the supine position but dropped markedly when sitting erect.

As the toxemia passed off the upper margin of liver dulness descended to the sixth rib, under about the same conditions of abdominal distention.

It is obvious from this example that marked variations in lung expansion might passively follow active changes in diaphragmatic tonus, assuming that such changes in tonus actually occur.

FURTHER OBSERVATIONS ON THE BEHAVIOR OF DIPHTHERIA BACILLI IN WHOLE COAGULABLE BLOOD

WITH SPECIAL REFERENCE TO BACTERICIDAL IMMUNITY IN TOXIN-IMMUNIZED AND BACTERIN-IMMUNIZED ANIMALS

By SOLOMON SOLIS-COHEN, M.D., MYER SOLIS-COHEN, M.D.
AND
LOUIS S. BOROW,
PHILADELPHIA

(ABSTRACT)

The observations here reported, while not in themselves alone of much importance, have considerable significance as links in a chain. Last year our lamented colleague, Dr. George D. Heist, reported to this Association[1] the result of our studies concerning the possible existence in the whole, coagulable blood of certain men and animals, of factors inimical to the growth and life of diphtheria bacilli, just as we (and others in confirmation) had previously demonstrated such factors with regard to the pneumococcus, the meningococcus and other organisms. Comparisons were also made between the results of the Schick (diphtheria toxin) test for antitoxic immunity and those of the Heist (whole, fresh, coagulable blood) test for bactericidal immunity upon the same individuals.

Briefly, it was shown that diphtheria bacilli could not be grown,

[1] Solis-Cohen, Solomon, Heist, George D., and Solis-Cohen, Myer: Observations on the Behavior of Diphtheria Bacilli in Whole Coagulable Blood, with a Comparison of the Results of Tests for Bactericidal and Antitoxic Immunity in the Same Persons. Tr. Assn. Am. Phys., 1920, xxxv, 263.

or grew but feebly, in the whole coagulable blood of an animal (the rat) exhibiting a high degree of original resistance to diphtheria; while they grew abundantly under like conditions in the whole coagulable blood of the guinea-pig, the mouse and the rabbit, as well as in the defibrinated blood, not only of these species and of the steer, but also of the rat. That the bacilli used were virulent toxin producers was shown by inoculation experiments, each of the four strains tested killing a guinea-pig within twenty-four hours.

As regards human beings, it was shown that the whole coagulable blood, both of certain adults and of certain children, killed or prevented the growth of virulent diphtheria bacilli, while that of other adults and other children did not; but that no correspondence could be established between the presence or absence of bactericidal immunity, as thus demonstrated, and the presence or absence of antitoxic immunity, as shown by negative or positive response to the Schick test. Sometimes both forms of immunity would be present in the same individual; sometimes both forms would be absent; sometimes one would be present and the other absent. There were both individual correspondences and individual divergences. Nor did the result seem to be influenced in any way by the fact of recovery from diphtheria some years earlier.

According to Park, recovery from diphtheria when antitoxin has not been administered may be attributed to the presence of bactericidal substances and not to that of antitoxin, since most of those who thus recover continue to show a positive Schick reaction. This is in accord with our findings. We have not yet been able to make comparative studies upon children recently recovered from diphtheria, but that should be done.

In order to determine whether active antitoxin immunization in animals is or is not accompanied by the development of bactericidal immunity, we tested the bactericidal power of the blood of horses possessing a high degree of antitoxic immunity. The whole coagulable blood, the defibrinated blood and the citrated blood were tested with three strains of B. diphtheriæ, two of them toxin producers and one a non-toxin producer. Absence of bacteria in sufficient numbers to be detected on the slide is taken to indicate

the presence of bactericidal power in the blood tested—but only with regard to that particular bacterium—and the presence of bacteria is held to indicate lack or low degree of such specific bactericidal power. The degree of specific bactericidal power, if any, is evaluated by the lowest dilution in which growth of the bacteria is prevented.

TABLE I.—ANTIBACTERICIDAL IMMUNITY IN TOXIN-IMMUNIZED HORSES

Tests made with:	Dilutions of cultures of Klebs-Löffler bacilli.				
	1 : 10	1 : 100	1 : 1000	1 : 10,000	1 : 100,000
Horse No. 1 (Serum Contained 300 Units Antitoxin per Cubic Centimeter)					
Whole coagulable blood:					
Non-toxin producer	++[1]	+[2]	±[2]	+[2]	+
Toxin producer, 161.4	+	−	+	±	−
Toxin producer, 194.4	+	+	±	±	−
Defibrinated blood:					
Non-toxin producer	++	++	++	++	0
Toxin producer, 161.4	++	++	+	+	+
Toxin producer, 194.4	++	+	+	+	+
Citrated blood:					
Non-toxin producer	++	+	+	+	+
Toxin producer, 161.4	+	+	+	+	+
Toxin producer, 194.4	+	+	+	+	−
Horse No. 2 (Serum Contained 350 Units Antitoxin per Cubic Centimeter)					
Whole coagulable blood:					
Non-toxin producer	++	++	+	+	++
Toxin producer, 161.4	++	++	++	+	+
Toxin producer, 194.4	+	+	+	+	+
Defibrinated blood:					
Non-toxin producer	++	++	++	+	+
Toxin producer, 161.4	++	++	++	+	+
Toxin producer, 194.4	++	++	++	++	++
Citrated blood:					
Non-toxin producer	+	+	+	+	+
Toxin producer, 161.4	++	+	+	−	−
Toxin producer, 194.4	++	+	+	+	+
Horse No. 3. Normal Control					
Whole coagulable blood:					
Non-toxin producer	++	++	+	−	+
Toxin producer, 161.4	++	++	±	+	+
Toxin producer, 194.4	++	++	++	++	++
Defibrinated blood:					
Non-toxin producer	++	++	++	++	++
Toxin producer, 161.4	++	++	++	++	++
Toxin producer, 194.4	++	++	++	++	++
Citrated blood:					
Non-toxin producer	++	++	++	+	+
Toxin producer, 161.4	++	++	++	++	++
Toxin producer, 191.4	++	++	++	++	+

[1] − indicates no growth; ± indicates doubtful growth; + indicates moderate growth; ++ indicates vigorous growth.

[2] Most forms took the stain poorly and were indistinct.

In a horse possessing 300 units of antitoxin in the cubic centimeter of serum there was, perhaps, evidence of a very slight bactericidal power in the whole coagulable blood, but none in the defibrinated or citrated blood. In the second horse, which had 350 units of antitoxin in the cubic centimeter of serum, no evidence of bactericidal action was observed either in the whole blood, the defibrinated blood or the citrated blood; and the same is to be said of a third (unimmunized) horse used as a control. One may therefore conclude that development or increase of antitoxic power in the blood through immunization by toxin, is not necessarily accompanied with the development of antibacterial properties.

It seemed desirable to determine, further, whether or not, in animals whose whole coagulable blood gave no evidence of power to restrain the growth of diphtheria bacilli, such property could be induced by the injection of diphtheria bacillary bodies in increasing doses. Two rabbits were injected intravenously with killed diphtheria bacilli at four-day and five-day intervals, the first dose being 500,000,000, the second dose 750,000,000 and subsequent doses 1,000,000,000, except in one case, in which increase was made to 1,250,000,000. The bactericidal power of the whole coagulable blood of each rabbit was tested before beginning the injections, and at intervals during their course, with living cultures of the same strain that was used for the attempt at immunization. In one the agglutinative titer of the blood serum was also taken.

In both rabbits the whole coagulable blood showed some slight development of bactericidal power. In one the agglutinative titer was 1:200 while in the normal rabbit it was 1:5. The other rabbit developed an acute paralysis of the hind legs and bladder —a not uncommon result of like experiments—and died.

The observations have not yet progressed sufficiently to permit of definite conclusions. If the bactericidal power can be sufficiently increased to indicate a possibility of a definite result the rabbits will be injected with living virulent diphtheria bacilli. This will determine whether or not such bactericidal power in the whole blood is accompanied by an immunity that will protect against diphtheritic infection. If sufficient degree of bactericidal power cannot be induced, some other animal should be studied in the same way.

TABLE II.—ANTIBACTERIAL IMMUNITY IN BACTERIN-IMMUNIZED RABBITS

RABBIT No. 1

April 9. The whole coagulable blood of the rabbit was tested with a virulent living culture of Klebs-Löffler bacillus.

	Dilutions of virulent living culture of Klebs-Löffler bacillus.				
	1 : 10	1 : 100	1 : 1000	1 : 10,000	1 : 100,000
Rabbit No. 1 . . .	++	++	++	+	+
Control	++	++	++	++	+

April 9. The rabbit was injected intravenously with 500,000,000 of killed Klebs-Löffler bacilli of the same strain.

April 14. The rabbit was injected intravenously with 750,000,000 of killed Klebs-Löffler bacilli.

April 18. The rabbit was injected intravenously with 1,000,000,000 killed Klebs-Löffler bacilli.

April 21. The whole coagulable blood of the rabbit was tested with a virulent living culture of the same strain of Klebs-Löffler bacillus.

	Dilutions of virulent living culture of Klebs-Löffler bacillus.				
	1 : 10	1 : 100	1 : 1000	1 : 10,000	1 : 100,000
Rabbit No. 1 . . .	++	++	+	+	+
Control	++	++	++	++	+

April 22. The rabbit was injected intravenously with 1,000,000,000 killed Klebs-Löffler bacilli.

April 26. The rabbit was injected intravenously with 1,000,000,000 killed Klebs-Löffler bacilli.

April 29. The rabbit's whole coagulable blood was tested with a virulent living culture of the same strain of Klebs-Löffler bacillus.

	Dilutions of virulent living culture of Klebs-Löffler bacillus.				
	1 : 10	1 : 100	1 : 1000	1 : 10,000	1 : 100,000
Rabbit No. 1 . . .	+	+	+	+	−
Control	++	++	++	+	+

April 30. The serum of rabbit No. 1 agglutinated 1:200. The serum of the control rabbit agglutinated 1:5.

RABBIT No. 2

March 10. The rabbit was injected intravenously with 250,000,000 killed Klebs-Löffler bacilli.

March 10. The rabbit's whole coagulable blood was tested with virulent living Klebs-Löffler bacilli of the same strain.

	Dilutions of virulent living culture of Klebs-Löffler bacillus.				
	1 : 10	1 : 100	1 : 1000	1 : 10,000	1 : 100,000
Rabbit No. 2 . . .	++	++	++	++	++
Control	++	++	++	++	+

March 15. The rabbit was injected intravenously with 500,000,000 killed Klebs-Löffler bacilli.

March 20. The rabbit was injected intravenously with 750,000,000 killed Klebs-Löffler bacilli.

March 25. The rabbit was injected intravenously with 1,000,000,000 killed Klebs-Löffler bacilli.

March 29. The rabbit's whole coagulable blood was tested with virulent living Klebs-Löffler bacilli of the same strain.

	Dilutions of virulent living culture of Klebs-Löffler bacillus.				
	1 : 10	1 : 100	1 : 1000	1 : 10,000	1 : 100,000
Rabbit No. 2 . . .	++	++	+	+	+
Control	++	++	++	+	+

March 30. The rabbit was injected with 1,000,000,000 killed Klebs-Löffler bacilli.

April 4. The rabbit was injected with 1,250,000,000 killed Klebs-Löffler bacilli.

April 8. The whole coagulable blood of the rabbit was tested with virulent living Klebs-Löffler bacilli of the same strain.

	Dilutions of virulent living culture of Klebs-Löffler Bacillus.				
	1 : 10	1 : 100	1 : 1000	1 : 10,000	1 : 100,000
Rabbit No. 2 . . .	+	+	+	−	−
Control	++	++	++	++	+

Rabbit No. 2 developed an acute paralysis of the hind legs and bladder and died.

The experiments here reported were conducted at the Jules E. Mastbaum Research Laboratory of the Jewish Hospital of Philadelphia.

DISCUSSION

Dr. William H. Park, New York: It occurred to me some years ago that there might have been an error in thinking that antitoxin immunity regularly caused a cure in diphtheria. We looked for children who had not received antitoxin, and discovered some in the country. We found that antitoxin did not occur in them until weeks after the recovery. In these it is probable that bactericidal immunity caused recovery. I think Dr. Cohen has chosen a most interesting and difficult field for investigation. There is no reason to fear that a single diphtheria antitoxin will not neutralize the toxin formed by all types. We have tried the toxins produced by hundreds of bacilli and have never failed to neutralize them by the standard antitoxin.

Dr. S. Solis-Cohen, Philadelphia (closing): My co-workers and I are fully alive to the importance of the questions raised by Dr. Park, and should be glad of his help in solving them. Dr. Park kindly allowed one of us to work on certain phases of the general problem in his laboratory. At that time no difference—or at least no significant time difference—could be shown between the growth in guinea-pigs' whole coagulable blood of a toxin-producing strain of diphtheria bacilli and that of two so-called non-toxin producers given us by Dr. Park. Subsequent work with these same strains, however, made us question whether the latter—that is to say the so-called non-toxin producers—should not rather be regarded as weak toxin producers. My co-workers recently grew in guinea-pigs' whole coagu-

lable blood, eighty-eight strains of diphtheria bacilli and organisms resembling these, furnished by the hygienic laboratory of the Bureau of Health of Philadelphia. The time at which growth was first manifested in the tubes was carefully noted; that is to say, whether this was before or after eight hours' incubation. At the same time inoculations were made with each strain on two guinea-pigs. One of each pair of animals was unprotected. The other, as a control, had received 500 units of antitoxin. Fourteen strains failed to show growth within eight hours and were non-pathogenic for both sets of guinea-pigs. These were regarded as Bacilli hoffmani. Seventy four strains showed growth at various periods within eight hours. Of these sixty-five strains killed only the unprotected guinea-pigs. They were regarded as virulent Klebs-Löffler bacilli. Six strains killed both the unprotected and the protected guinea-pigs and were regarded as identical with those bacilli commonly found at autopsy in the blood and internal organs. Three strains produced a transient illness in the unprotected guinea-pigs from which the control guinea-pigs were slightly protected. These were regarded as Klebs-Löffler bacilli that were slight or weak toxin producers. It is the problem of this last group—the weak toxin producers—that particularly requires further study. We fully realize the difficulties of the next steps and would welcome the coöperation of other workers to confirm, correct or extend our observations.

YAWS: AN ANALYSIS OF 1046 CASES

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At the request of the military government of the Dominican Republic the School of Tropical Medicine of Harvard University sent a commission, consisting of Drs. A. W. Sellards, W. L. Moss and G. H. Bigelow, to Santo Domingo during the summer of 1920 to study yaws. The results of the observations made are herewith presented in abstract.

As the disease is strictly limited to tropical and subtropical countries it may be unfamiliar to many physicians who have not been in the tropics. Therefore a brief introductory description of yaws is given.[1]

Synonyms. Frambesia tropica, yaws, buba, pian, etc.

Definition. A tropical, specific, infectious and contagious disease caused by Treponema pertenue Castellani and characterized by a frambesiform granulomatous eruption.

Distribution. West Indies, Central America, northern part of South America, Africa, Malay Peninsula, Siam, Ceylon and parts of China. It is said never to occur in the mountains and cold districts and rarely above an elevation of 800 feet.

Etiology. The Treponema pertenue, first observed by Castellani in 1905, is now accepted as the etiologic agent of the disease. There has been much discussion concerning the question of the identity of yaws and syphilis, and while some observers still main-

[1] This description is taken largely from the chapter on Frambesia Tropica (Castellani and Chalmers: Manual of Tropical Diseases, third edition, William Wood & Co., 1920).

tain the unity of the two diseases the evidence at hand seems sufficient to establish them as separate diseases. Charlouis, as early as 1882, successfully inoculated yaws in a syphilitic patient and syphilis in a patient infected with yaws. That yaws patients are not immune against syphilis is further indicated by the observations of Powell and Nichols and others, who have described cases of syphilis supervening on yaws.

Incubation Period. Castellani gives the incubation period as two to four weeks. Paulet, in 1848, inoculated 14 negroes with the secretions from frambetic granulomata and all developed yaws in ten to twenty days. In monkeys and other animals experimentally inoculated with yaws the incubation period varied from sixteen to ninety-two days.

Symptomatology. The course of the disease, like syphilis, may be divided into a primary, secondary and tertiary stage. Some authors add a fourth stage (paraframbesial affections), but it is questionable if the observations on which this additional stage is postulated are reliable.

Primary Stage. Incubation period two to four weeks, characterized by malaise, rheumatic pains, headache and sometimes irregular fever. The primary lesion (mother yaw, madre buba) appears as a papule, which after a week becomes moist and develops a yellow secretion. A crust may form, which on removal reveals an ulcer with a granulomatous base. The primary lesion may heal before the general eruption appears, but usually persists until after the secondary eruption sets in.

Secondary Stage. The secondary stage begins one to three months after the primary lesion and is characterized by a general eruption of granulomata over the body. It is preceded by malaise, headache, severe pains in the muscles, joints and bones. In some cases there may be fever of an intermittent type. The granulomata appear first as minute papules, which increase in size to an average diameter of about 1 cm. They may coalesce, become secondarily infected and form large ulcerating areas. In the majority of cases—within three to six months in children and six to twelve months in adults—the granulomata dry up, shrink and disappear. In some cases the granulomatous eruption may continue for several

years, new crops of nodules appearing from time to time in succession. The granulomata generally undergo involution within two to four months, leaving behind, as a rule, some change in pigmentation. At times secondary ulceration occurs, leading to deeper scars, and occasionally the granulomata take the form of a circinate eruption.

The granulomata frequently occur on the soles of the feet, less often on the palms of the hands, resulting in a condition closely resembling the syphilitic psoriasis palmaris and plantaris.

The alimentary, respiratory, excretory and central nervous systems are rarely involved. The joints are frequently swollen and painful. Lesions of the mucosæ are rare. Castellani and Chalmers have not observed allopecia, but state that various groups of lymphatic glands are found enlarged. There may be a moderate degree of anemia of secondary type and an increase in the large mononuclears. The Wassermann reaction is positive in the great majority of recent cases, but is fairly often negative in old cases.

Tertiary Stage. The disease often terminates with the second stage. Sometimes the secondary and tertiary stages merge or there may be an interval of years between them.

The characteristic lesions of the tertiary stage are gummatous-like nodules and deep ulcerative processes, frequently resulting in great deformity. Bone lesions are common, but visceral lesions are rare or absent. Castellani and Chalmers think that gangosa, an ulcerative condition of the palate, nose and pharynx, is in reality a tertiary manifestation of yaws.

Histopathology. In the frambetic papules the surface epithelium is greatly thickened and numerous elongated downgrowths are seen. In patches the epithelial cells are swollen, vacuolated and degenerating. Circumscribed areas contain polymorphonuclear leukocytes. The layers near the corium are almost normal. The corium itself is edematous. There is a diffuse cellular infiltration consisting of polymorphonuclear leukocytes, large and small mononuclears, eosinophils, plasma cells, mast cells, connective-tissue cells and some extravasated erythrocytes. Treponemata are present in the granulomata.

Treatment. Brodin, in 1910, tried antimony.

"Castellani's Yaws Mixture" contains tartar emetic, potassium iodide, sodium salicylate and sodium bicarbonate.

Salvarsan was first used by Nichols in the experimental disease in monkeys and by Strong in the treatment of the disease in humans.

The cure of yaws by salvarsan or neosalvarsan is one of the most striking examples of specific therapy that medicine presents.

The malady does not appear to be hereditary. The course of the disease is chronic, resulting in serious disability and much suffering in a majority of cases. The mortality from yaws is not high, and death, when it occurs, is usually due to secondary infections.

AUTHOR'S OBSERVATIONS

In discussing the cases observed by us in Santo Domingo the following abbreviations are used for convenience of reference. If the reader will take a moment to familiarize himself with these the understanding of what follows will be facilitated.

TABLE OF ABBREVIATIONS.

M	Madre buba, mother yaw. Primary lesion.
B′	Florid secondary eruption of granulomata. Early secondary stage.
B	Sparse, recurring, secondary granulomata. Late secondary stage.
C	Clavus. Late lesions on the soles. Late secondary stage.
P	Palmar lesions. Late lesions on the palms. Late secondary stage.
G	"Gomma." Tertiary stage.
H	History of yaws. No active lesions present. Quiescent.

Primary Stage. We observed 64 cases in which the primary lesion was present, sometimes alone, oftener in combination with other manifestations of the disease. Briefly these cases may be presented in the following tabular form:

Diagnosis.	No. of cases.	Average age yrs.	Average duration, mos.
M	15	6.6	2.3
MB′	25	8.5	7.3
MB′C	13	11.6	4.1
MB	6	24.2	19.0
MBC	5	10.0	17.2

The extremes of age in this group of cases were seven months and sixty years. The duration of the madre at the time of observation

varied from one month to six years, and the size varied from 1 to 7 cm., with an average diameter of about 2.5 cm. Of the 15 cases in the above group which presented a madre buba as the only active lesion, 5 showed healed scars of secondary granulomata. Thus there were only 10 cases which had not progressed beyond the primary stage. In 54 cases, or 84 per cent of our series, the madre persisted after the appearance of secondary lesions.

Location of Primary Lesion. In 969 cases the location of the madre was determined either by history, observation of the lesion or scar (in 635 cases, or 71 per cent, the scar was present). In 18 additional cases patients stated that they had had no madre but that multiple granulomata appeared simultaneously over the body. In 2 cases patients claimed to have had two madres simultaneously.

TABLE SHOWING LOCATION OF MADRE.

	Cases.	Per cent.
Lower extremities	803	82.86
Upper extremities	80	8.25
Head	39	4.02
Trunk	37	3.81
Genitalia	10	1.03
	969	99.97

The primary lesion occurred on the right side of the body in 425 cases and on the left side in 468 cases. In the remaining cases the site was noted as the knee, the ankle, etc., but the side of the body was not recorded.

These figures have been analyzed to determine if the site of the madre might throw any light on the mode of transmission of the disease. In this connection it is to be remembered that we are dealing with a barefooted and barelegged population; that even the adults are scantily clothed; that the upper extremities are almost as much exposed as the lower, and that children of both sexes usually wear no clothing until they are about five years of age.

Sexual Transmission. Sexual transmission as a common method may be excluded because of the fact that in only 1 per cent of our series was the primary lesion on the genitalia. Of these 10 individuals 6 were males, 4 females and 5 were under eight years of age.

Moreover, in 25 per cent of all the cases in our series the infection occurred before the fifth year of life.

Hereditary Transmission. We saw no evidence of hereditary transmission in any case.

Insect Transmission. We think that transmission by the bites of the common flying insects, such as flies, mosquitoes, etc., may be excluded on the basis of the distribution of the primary lesion: In the great majority of cases the madre occurred on the lower extremities, whereas the face and upper extremities are equally exposed to the bites of these insects.

The preponderance of the primary lesion on the lower extremities and over the trunk would likewise seem to exclude transmission by the bites of such non-flying insects as lice, fleas, bed bugs, etc.

The fact that in such a large majority of cases (83 per cent) the primary lesion occurred on the lower extremities led to a further analysis of these cases. The result is shown in the following tabulation, and for comparison the figures for the upper extremities are given:

Buttocks	13 cases	Shoulders	6 cases
Thighs	30 "	Upper arms	4 "
Knees	55 "	Elbows	16 "
Legs	201 "	Forearms	17 "
Ankles	258 "	Wrists	8 "
Feet	246 "	Hands	25 "
Total	803 cases		76 cases

It will be noticed that the frequency with which the primary lesion is found on the various parts of the lower extremities increases strikingly as one approaches the ground. This suggests that if the disease is transmitted by the bites of insects we should think of some non-flying insect which remains on or near the ground and does not normally prey on man or even desire to domicile with him—such as ants, ground spiders, etc.

The possibility of trauma forming the portal of entry for the treponema has been suggested. The greater frequency of the primary lesion on the exposed parts of the body fits in with such an hypothesis. It seems probable also that in a barefooted population the lower extremities would be the most frequently trauma-

tized part of the body and that the frequency of minor injury would increase as one progressed down the extremity. Examination of the preceding table shows that the frequency with which the madre occurs on the various parts of the lower extremities increases as one descends toward the foot.

Man being a forward-going animal is liable to get most of his knocks from in front. Accordingly we have analyzed the primary lesions occurring on the knees, legs and ankles with reference to their occurrence on the anterior, external, internal and posterior surfaces.

As far as our records showed this data the results are set forth in the following table:

	Anterior.	External.	Internal.	Posterior.
Knees	18	8	6	4
Legs	107	35	19	12
Ankles	25	79	84	13

In the case of the knees and legs it is shown that the anterior surface leads by a large majority, the external surface comes next, followed by the internal surface, while the posterior surface finishes a poor last. This probably corresponds closely to the exposure to minor traumata.

In regard to the ankle the circumstances are different. Owing to its conformity the malleoli are about equally exposed to injury; while the anterior surface of the ankle is protected by the projecting foot, which receives on its dorsum the injuries which would otherwise go to the ankle. This corresponds to the frequency of the madre on the various parts of the ankle, and to the fact that in case of the foot the primary lesion was found on the dorsum in a great majority of cases.

It appears probable from our observation, therefore, that trauma, frequently minor in character, usually determines the site of the primary lesion in yaws. This is entirely compatible with the view that the disease may be transmitted by direct contact or that insects, flies, etc., may be vectors of the virus.

Secondary Stage. *Florid Secondary Stage.* In our series there were 144 cases who presented themselves during the florid secondary stage of the disease. In 38 of these the primary lesion was still

present and in 37 the late lesions of the soles, known as clavus, had made their appearance.

Briefly summarized these cases are presented in the following table:

FLORID SECONDARY STAGE

Diagnosis.	No. of cases.	Average age, yrs.	Average duration, mos.	No. cases with fresh granulomata on:	
				Soles.	Palms.
MB′	25	8.5	7.3	2	1
MB′C	13	11.6	4.1	7	1
B′	82	8.8	12.3	6	0
B′C	24	12.9	30.4	9	2
	144			24	4
				16.6 per cent	2.8 per cent

Exclusive of the 38 cases still showing the primary lesion and previously analyzed the youngest patient in this group was one year and the oldest fifty years.

While clavus may develop very early in the disease—witness the 13 MB′C cases with an average duration of four and one-tenth months—as a rule it does not develop until the disease has lasted more than a year. The average duration of the 82 cases which presented only secondary granulomata was twelve and three-tenth months, while the average duration of the cases which had clavus in addition to the florid secondary eruption was thirty and four-tenth months. The greater frequency of granulomata on the soles as compared with the palms is indicated by their presence in the former situation in 16.6 per cent of this group as against 2.8 per cent in the latter situation.

From our observation and the histories we obtained we are led to believe that in the disease as it occurs in Santo Domingo practically all of the patients, sooner or later, develop granulomata on the soles and become the victims of the painful condition which they designate as clavus.

The secondary eruption as we saw it corresponds closely to the text-book description as given by Castellani and Chalmers. The individual granuloma does not differ materially in appearance from the primary lesion except that it usually does not become as large and seems less liable to undergo as deep ulceration. In our notes we described the earliest stage that we observed as a small papule

1 to 2 mm. in diameter, gray in color and surrounded by a narrow zone of hyperemia. These papules increase rapidly in size to an average diameter of about 1 cm., and are usually elevated $\frac{1}{2}$ to $\frac{3}{4}$ cm. above the skin. The surface of the granuloma is covered with minute bosses which give it a raspberry-like appearance from which the disease takes the name frambesia. Early the granulomata are of a pearly gray color, later they are liable to become covered by a thin yellow crust. After a few weeks or months retrogressive changes take place: the lesions turn dark, often black in color, shrink in size, the surface becomes checked, and after a time all that remains is a black crust, which on falling may leave no scar; more frequently pigmentary changes persist, which may consist of increased pigmentation, apigmentation or an apigmented area surrounded by a zone of increased pigmentation.

At no stage of the granulomata was vesiculation noted. They may go through their entire existence without ulceration, but more frequently they become secondarily infected and consequently more or less deeply ulcerated. During the florid secondary eruption there may be hundreds of granulomata over the body and no part of the cutaneous surface, from the crown of the head to the soles of the feet, is exempt from invasion. They show a special predilection for the mucocutaneous borders—nostrils, mouth, genitalia, anus and moist surfaces; axillæ, internatal region, between the upper thighs and popliteal spaces. The mucous membranes are said to be rarely involved and we saw but one such case in our series, a single granuloma on the vermilion surface of the lower lip. We observed no case of allopecia except as the result of ulcerative processes involving the scalp. One patient presented the typical circinate arrangement of the lesions on the face, which strikingly suggested the Indian war-paint appearance.

Usually there was no complaint of pain in consequence of granulomata on the body except in cases in which they were situated on the bearing surfaces of the feet, on the palms of the hands, or when extensively ulcerated.

Arthritis and dactylitis were fairly common, although they usually occurred later than the florid secondary stage of the disease. The knee-joints were most frequently involved; the elbows were

affected next in frequency. Usually a single joint was affected. The swelling was sometimes considerable and in one instance as much as 200 cc of slightly turbid, straw-colored fluid was aspirated from a knee-joint. This fluid was without evident effect when injected into the peritoneal cavity of a guinea-pig. The dactylitis took the form of the usual spindle-shaped fingers, generally a single digit, sometimes several, being involved.

Late Secondary Stage. There were 190 cases in the series that were considered to be in the late secondary stage of the disease. They had passed through the florid secondary eruption, as evidenced by the numerous scars of previous granulomata. Many of them had doubtless been entirely free from active lesions for variable lengths of time when one or more recurrent granulomata appeared, it is evident from the histories obtained that the disease may undergo an apparent spontaneous cure, but sooner or later granulomata reappear in a majority of cases and successive crops, consisting usually of only a few lesions, continue to make their appearance, with free intervals between, for many years. Briefly tabulated these cases may be presented as follows:

LATE SECONDARY STAGE

Diagnosis.	No. of cases.	Average age, yrs.	Average duration.	No. cases with granulomata on: Soles.	Palms.
MB	6	24.2	19.0 mos.	2	1
MBC	5	10.0	17.2 "	1	1
B	60	9.3	29.6 "	16	1
BC	106	16.5	5.4 years	69	1
BCP	13	20.5	11.3 "	10	4
	190			98	8
				51.6 per cent	4.2 per cent

Of the 190 cases in this group, 60 had as their only active manifestation of the disease a few secondary granulomata, 106 cases had granulomata plus clavus lesions and 13 cases had in addition to the above palmar lesions; 11 cases which have already been analyzed with the madre cases are included here, as they appear to have been in a late secondary stage of the disease in spite of the persistence of the primary lesions.

The extremes of age in this group are one and seventy-five years; and while the former figure shows that a late secondary stage may

be reached not only early in life, but comparatively early in the disease, the average age in this group indicates that this is exceptional, and both the average age and average duration of the disease at the time of observation point to the chronicity of the secondary stage of the disease.

Clavus. Perhaps the most curious and interesting effects of the disease are the late lesions which occur on the soles of the feet, and it is to these that much of the pain and disability from the disease are due. We have adopted the term "clavus," used by natives, because it seems the appropriate one, although they use it to designate conditions of the soles long after anything suggesting a "nail" has disappeared.

In a previous section attention has been called to the frequency with which secondary granulomata appear on the soles. Those occurring in this situation do not differ materially in appearance from granulomata occurring elsewhere on the body. They are perhaps more frequently secondarily infected and are liable to ulcerate, probably as a result of the fact that the natives seldom protect them by any sort of dressing, and continue to hobble about even when the feet appear to be in a shocking condition.

If the granuloma on the sole heals without ulceration, as it shrinks it separates from the surrounding epidermis and becomes circumvallate. The hard, dry, black core must act like a foreign body beneath the foot, and it is the "nail" which led to the use of the term clavus. Finally the core drops out, leaving the "nail hole," a circular opening $\frac{3}{4}$ to 1 cm. in diameter, with sharp-cut edges which descend vertically 2 or 3 mm. to a flat base.

Usually both soles are about equally involved, although rarely cases are encountered in which only one sole is affected. The number of granulomata on each sole may vary from only a few to 20 or 30. Irregular erosion, the result of attrition of the epidermis intervening between the "nail holes," may account for the remarkable motheaten appearance of the soles which many of the old clavus cases present.

Under the designation "clavus" the natives included conditions differing from those just described and which are not so readily explained as the result of preëxisting granulomata. Not infrequently the soles presented a condition which is aptly described

by the term honeycombed. The entire sole or, in some cases, only the bearing surface was filled with pits ½ cm. or less in diameter, conical in shape and extending to a depth of 2 or 3 mm. These pits are as closely and regularly set as the cells of a honeycomb, and no such number or distribution of fresh granulomata was observed as would suggest that each pit could represent the site of a previous tumor.

In other cases the epidermis of the entire sole or a large part of it was greatly thickened, black and dead looking.

The natives applied the term clavus indiscriminately to any of the conditions just described. Pain was the characteristic common to all. Like the granulomata elsewhere on the body, the clavus lesions would clear up spontaneously in time, only to recur again and again throughout many years.

Occasionally a condition of the soles was seen, to which the natives apply the name "rajadura," which means a fissure or crack. The fissures, 2 or 3 cm. long and 2 or 3 mm. deep, are usually arranged vertically about the periphery of the heel. More rarely they occurred in the sole beneath the instep or ball of the foot, where they might attain a length of 6 to 8 cm., reaching to a considerable depth, and, like those of the heel, cause much pain. We gained the impression that these fissures were also due to yaws, and for this reason have included in our analysis the relatively small number of cases encountered with clavus, although the term "rajadura" is more descriptive.

Before proceeding to the analysis of the clavus cases it is well to state that we have not included in this category any cases which presented fresh granulomata as the only lesions of the soles. We have reserved the term clavus to designate only the late effects of the infection on the soles, and think that these should be regarded as belonging to the late secondary stage of the disease. It may be of interest to give here a description of the changes which took place in the clavus cases following the administration of neosalvarsan leading to their cure. A statistical statement of the result of the treatment in all cases will be given in a later section of this paper, but the results obtained by treatment furnished corroborative evidence that these several different lesions were properly considered as a manifestation of yaws.

We were prepared to expect the improvement which resulted

from the use of neosalvarsan in the cases with fresh granulomata, whether they were situated on the soles or elsewhere on the body; but it seemed rather too much to expect that these old "motheaten," dilapidated soles would be rejuvenated by a few injections of any drug. The first few cases were given an injection more as a *placebo* on account of the importunity of the patients and the long distances that some of them had come rather than with any expectation that they would be benefited. When these patients came back at the end of a week, however, their feet not showing the least improvement in appearance, we received with surprise and incredulity the statement that they were cured of pain, and when the patients brought forth in support of their claim the statement that whereas they were obliged to come on horse the first time they were able to return on foot, we were encouraged to try treatment on other cases. In the end clavus comprised the largest and in certain respects the most important group of cases that presented itself for treatment.

A week after the first injection of neosalvarsan there was, as a rule, no objective change in the feet, but almost without exception the patients affirmed that the pain was either greatly ameliorated or entirely gone. The injections were usually given at seven- to ten-day intervals. About a week after the second injection the old dead epidermis loosens up about the edges of the clavus holes, erosions and fissures, and begins to desquamate. This process goes forward rapidly, until within a week or two after the second injection all the old epidermis has been shed, exposing healthy pink skin.

The following table summarizes the clavus cases:

Diagnosis.	No. of cases.	Average age, yrs.	Average duration.	Sole affected.		
				Right.	Left.	Both.
MB′C	13	11.6	4.1 mos.	8	11	6
MBC	5	10.0	17.2 "	4	4	3
B′C	24	12.9	30.4 "	21	23	20
BC	106	16.5	5.4 years	94	92	80
BCP	13	20.5	11.3 "	11	13	11
C	327	21.6	9.0 "	299	291	266
C+[1]	41	29.4	12.9 "	33	35	27
CP	50	32.7	18.7 "	45	47	42
	579			515	516	455

[1] The group of 41 cases designated as C+ had in addition to clavus some other condition which apparently was not dependent on the yaws infection. In a majority of cases the additional affection consisted of chronic leg ulcers.

In 3 of the 579 clavus cases no note was made as to which sole was involved. It is apparent, however, that the right and the left sole are affected with equal frequency and that in a majority of cases both soles are involved (79 per cent in the above series).

Although we have seen clavus in an infant, aged one year, attention is called to the fact that the average age of the largest group in this series, the 327 patients who had clavus only, was twenty-one and six-tenth years, and that the average duration[1] of the disease in this group at the time they presented themselves for treatment was nine years.

Palmar Lesions. Perhaps corresponding to the clavus lesions, though not so definitely dependent upon granulomata, are the painful keratoses and fissures which, late in the disease, appear in the palms of the hands. There is sometimes more or less erosion of the epidermis and frequently an inability to open the hand fully and extend the fingers. This is apparently due to a certain amount of contraction of the hard, dry, keratosed surface.

The condition is usually painful, though less so than clavus, and interferes with manual labor. It is a much rarer lesion than clavus and appears to develop in older individuals and later in the disease, though it seems to be equally amenable to treatment.

In our series this condition was met with 68 times, but only 5 cases had palmar lesions (exclusive of scars) as the only effect present. These 68 cases are briefly summarized in the accompanying table:

Diagnosis.	No. of cases.	Average age.	Average duration.
BCP	13	20.5 years	11.3 years
CP	50	32.7	18.7
P	5	25.0	14.0

It is rather striking that of the cases presenting a single manifestation of yaws, clavus forms the largest group, 327 cases, while those having palmar lesions (other than fresh granulomata) constitute the smallest group, 5 cases. It is also striking in the above summary of the palmar cases that those associated with clavus as the only other lesion form the largest number, 50 out of 68 cases.

[1] Wherever average duration is given in this paper it refers to the duration since the first appearance of the primary lesion and not to the duration of any particular stage unless so specified.

It is interesting that the average duration of the disease in the CP group, eighteen and seven-tenth years, is the longest of any group of the entire series, not excepting those in the tertiary stage of the disease.

Studded Lesions. In a number of cases we observed nodular skin lesions which we believe to be a manifestation of yaws. The earliest stage of these nodules was either not observed or else was not recognized as the beginning of lesions which in a later stage of their development became quite familiar to us. Apparently these lesions may occur on any part of the body, although we never observed them on the head or face. The forearms and legs were the commonest sites in the cases which we observed. The thighs and trunk were frequently involved and lesions starting on the dorsum of the foot and extending on to the plantar surface were noted.

A typical picture of the condition as observed by us may be described as follows: The individual lesion consists of a moderately hard skin nodule, 1 cm. in diameter, elevated 3 or 4 mm., not painful, unaccompanied by itching and without striking pigmentary changes until after regression, when increase of pigment may mark its former site. These nodules are thickly studded and regularly set over an area 8 to 10 cm. in diameter. The size of the area involved increases by an advancing margin consisting of an almost unbroken row of nodules. As this peripheral advance occurs, healing takes place in the center of the area. If the nodules do not ulcerate this retrogression is accompanied by desquamation of the epithelium. The nodules gradually flatten out and finally disappear, leaving no trace or more often a circular area of increased pigmentation.

In no case were the nodules observed to pass through a vesicular stage, but they frequently underwent ulcerative changes varying from very superficial ulceration involving only the individual nodules to extensive and deep ulceration which sometimes became confluent over large areas of the body and extended to the subcutaneous tissues. The degree and character of the resultant scarring probably depends upon the depth to which the ulceration had extended. Sometimes there remains only an increase of pigmenta-

tion as mentioned above, sometimes the skin over the entire area involved was left thin and crinkly and in patterns like those which form on the surface of hot chocolate which has been thoroughly boiled and allowed to stand a while. Again, where the ulceration has extended to a greater depth there may result complete leukoderma and sometimes painful keloid.

We have called these lesions "studded" for the lack of a more descriptive term. The area involved when small is usually circular, but as it increases in size it is liable to become elliptical or more often it approaches a rectangular shape, but with rounded corners. If situated on an extremity the long diameter of the area coincides with the axis of the limb. Seldom does it completely encircle an extremity even when large areas are involved.

We regret that we had no opportunity of studying these lesions histologically. They do not in the least resemble the primary and secondary granulomata. We do not know if they should be considered a late secondary manifestation of the disease or if they belong to the tertiary stage.

We have notes on 37 cases, the youngest patient exhibiting these lesions being five years of age and the oldest eighty years. The average age in this group being twenty-eight and eight-tenth years, which comes next in length to the gomma group (twenty-nine and three-tenth years). The average duration of the disease (yaws) in this group was thirteen and five-tenth years with the extremes of duration five months and thirty-nine years. This average duration is exceeded only by the P, G and CP groups.

Our belief that these skin lesions are a manifestation of yaws is based on the fact that they were observed only in patients who gave a history of yaws, some of whom had other recognized yaws lesions, and on the observation that they apparently responded specifically to neosalvarsan therapy.

We are inclined to regard them as a late secondary manifestation of the disease.

Tertiary Stage. The impression gained from a fairly close study of over 1000 cases leaves no doubt in our minds that yaws and syphilis are separate diseases. Exclusive of the evidence contained in the literature of inoculation experiments, the occurrence

of yaws and syphilis in the same patient, the difference in the response of the two diseases to mercury, possibility of reinoculation in yaws, the localization of the disease to the tropics and below certain altitudes, the grounds afforded by our own experience on which we base our opinion are as follows: Syphilis is universally recognized as a venereal disease; the primary lesion is, in the vast majority of cases, located on the genitalia and even in the cases not contracted through sexual intercourse, in which the primary lesion is extragenital, the course of the disease is in no way modified by the method of inoculation or the unusual seat of the chancre. Yaws, as we have already pointed out, can be excluded from the category of venereal diseases by the fact that in a series of approximately 1000 cases the primary lesion was located on the genitalia in only 1 per cent and infection took place in 25 per cent before the fifth year of life. Moreover the primary lesion in yaws differs distinctly in appearance from chancre even when the latter is extragenitally located—the madre buba, as a rule, being larger, the surface rougher and in no case was there the hardness sometimes present and usually considered characteristic of the syphilitic lesion.

In no case did we observe any evidence or obtain any history of hereditary transmission of the disease.

The duration of the madre buba and the high percentage of cases in which it persists after the onset of the secondary stage of the disease (84 per cent in our group of 64 madre cases) is contrary to what is observed in syphilis.

When we come to consider the secondary stages of the two diseases we find more striking contrasts. The remarkable pleomorphism of the secondary lesions of syphilis and the faithful monotony of the secondary granulomata of yaws. The versatile Treponema pallida may occasionally counterfeit yaws, but never did we observe a yaws patient in the secondary stage of the disease who failed to show either typical granulomata or scars which we felt sure were the marks of typical granulomata. Even the secondary infections and ulcerations which may result do not disguise the nature of the lesions.

Although Castellani and Chalmers say that "occasionally peeling,

whitish patches may be seen on the palms of the hands and soles of the feet closely resembling the syphilitic psoriasis palmaris and plantaris," we have never observed in syphilis anything resembling the clavus and palmar lesions which we have described as occurring in yaws. On the other hand none of the yaws cases which we observed presented such characteristic secondary manifestations of syphilis as the macular skin rash, mucous patches and allopecia.

In the tertiary stage of the disease the absence of visceral involvement, especially of the heart, bloodvessels, liver and kidneys, is in striking contrast with the frequency of their involvement in syphilis. Moreover, in a series of over 1000 cases in which the average duration for the entire series was twenty and three-tenth years, 79 of which were in the tertiary stage, none showed any evidence of central nervous system involvement. We do not wish to attach undue significance to the absence of central nervous system lesions in this series of cases, owing to the relative immunity of the colored race to such involvement in syphilis.

Finally the marked difference in the response of yaws and syphilis to neosalvarsan is a not unimportant point in favor of the duality of the two diseases. We have seen the lesions of a patient in the florid secondary stage of the disease melt away completely after a single injection of 0.6 gram neosalvarsan. Indeed the cure of yaws by this agent is the most dramatic therapeutic performance that we have ever witnessed.

While the differential diagnosis between syphilis and yaws in the primary and secondary stages presented little difficulty, there were cases presenting tertiary lesions in which we found it impossible to make the differential diagnosis. There were patients with periosteal thickenings of the long bones, especially of the tibia, radius and ulna. There were those with typical spindle-shaped swellings of one or more fingers. Others with the bridge of the nose destroyed and with perforation of the hard and soft palate (the condition known as gangosa). There were cases in which old ulcerative processes had led to loss of substance of the cranial and other bones. Some of the cases in which an eye had ulcerated out, in which the nose was almost completely destroyed or fingers or toes had been lost suggested the ravages of leprosy. Whether these results were

due to syphilis or yaws we are unable to state positively. The patients themselves applied the term "gomma" to all tertiary lesions and stoutly maintained that they were due to the latter disease, yaws.

There were other patients exhibiting tertiary manifestations which bore no resemblance to syphilis as we see it in the United States today. These included extensive skin ulcerations, great enlargement and deformity of the hands, especially of the legs and feet.

As previously mentioned, pain in the joints and often an effusion was a frequent complaint in the secondary stage. Many of the tertiary cases showed arthritic enlargement and partial or complete fixation.

That those cases, objectively indistinguishable from syphilis, were actually yaws we must, of course, leave unsettled. In favor of the view that they were yaws are the following facts:

The patients themselves, who are familiar with both diseases, maintained that the condition from which they were suffering was yaws. Practically all of this group denied having had syphilis (this denial deserves some consideration because little or no shame is attached to venereal infection among the natives). There were 79 individuals in the "gomma" group, and of these 67 were able to give the location of the primary lesions. Corroborative evidence of the correctness of their statements as to the location of the primary lesion was furnished by a scar in 34 cases. In only 2 of the 67 cases was the location of the primary at all a usual one for a chancre—the upper lip in one case and the perineum in the other.

SUMMARY OF THE GOMMA CASES

Diagnosis.	No. of cases.	Age in years.		Duration in years.	
		Average.	Extremes.	Average.	Extreme.
G	67	29.3	3–85	16.3	7/12–59
MG	1				
BG	3				
BCG	2	25.2	7–60	9.7	1–40
CG	5				
CPG	1				

The youngest patient in the entire gomma group was three years and the oldest eighty-five years. The average age for the

entire group as well as the duration of the disease at the time of observation was greater, as would be expected, than in any other group. The extremes both of age and duration are strikingly far apart.

It is of interest that in one case the primary lesion persisted after tertiary manifestations had made their appearance. That the disease may progress to the tertiary stage in a comparatively short time is shown by the fact that 10 (or 7.9 per cent) of the gomma cases had had the disease two years or less at the time of observation.

Latent Yaws. We have included in our series 127 individuals who had no active lesions of yaws at the time they were observed, but who gave a history of having had the disease, and who, in most cases, exhibited confirmatory evidence in the way of scars. Brief histories were taken and examinations made of all these cases, as they furnished certain statistical data of some interest. Moreover, being impressed with the chronicity of the disease, the frequent recurrence of active lesions after longer or shorter periods of latency, doubting if spontaneous cure occurred in any considerable proportion of the cases, and at the earnest solicitation of these individuals, many of whom would say, "I had clavus last month and I will have it again next month," or "I have a breaking-out of the buba every year," we made a practice of giving one or more injections of neosalvarsan to cases in which the history and examination satisfied us that they probably had had yaws.

The 127 cases which come under this heading may be tabulated as follows:

Diagnosis.	No. of cases.	Age in years.		Duration in years.	
		Average.	Extremes.	Average.	Extremes.
H	55	21.2	1–80	8.8	2/12–50
H+	27	22.9	2–62	10.2	6/12–45
H (ulceration)	45	27.6	10–80	10.6	3/12–59

Of the cases in this group those designated H were without active lesions of any disease but gave a satisfactory history of having had yaws. Under H+ we have grouped those individuals who in addition to giving a history of having had yaws had symptoms or signs of some other disease, such as various skin eruptions, pains in various parts of the body, extensive scarring, etc. One

case gave a history of chancre and had a scar on the prepuce. The H (ulceration) group comprises 45 cases which, in addition to a history of yaws, had active ulceration on some part of the body, usually old leg ulcers, which we could not definitely identify as yaws and in most cases believed to be due to some other condition.

It seems not unlikely that this group of 127 individuals must have included many latent cases of yaws which, if left untreated, sooner or later would have again developed active lesions, and not only suffered the disability arising from the disease themselves but would also have become active agents in the spread of the disease. The statistics of this group seem to lend weight to this view. The average age in this group (twenty-three and eight-tenth years) is lower than the average age of various other groups which had active lesions, *e. g.*, Groups MB, C+, CP, P, G and G+. Moreover the average duration of this group (nine and seven-tenth years) is shorter than that in several groups having active lesions, *e. g.*, C+, CP, P and G.

Indeed in 33 (or 26 per cent) of these 127 cases the primary lesion had developed within two years prior to the time they applied for treatment. It seems highly probable that some of these 33 cases may have been in a free interval between the primary and secondary stages of the disease and that the majority of them were in a latent period following the early secondary eruption.

With a very limited time in which to work among a given population and with the object of accomplishing as much as possible toward eradicating the disease from the locality in which we were stationed, we think the policy of treating cases which gave a history of having had the disease, even though without active lesions at the time, is fully justified.

General Remarks. *The epitrochlear glands* were noted in 886 cases and were found to be palpably enlarged in 519 (or 58.5 per cent). They varied in size from a grain of wheat to 1 or 2 cm. Sometimes two or three glands 1 cm. in diameter were found in the epitrochlear region.

The femoral glands were noted in 121 cases and found enlarged in 100 per cent of the cases noted. In a majority of cases the femoral glands formed a visible ovoid swelling in Scarpa's triangle,

frequently attaining a width of 3 or 4 cm. and length of 5 or 6 cm. It seems probable the enlargement of this group of glands may have been due to the frequent pyogenic infections on the feet and legs that occur in a barefoot population.

The spleen was noted in 216 cases and was found enlarged in 58 (or 26.8 per cent). Malaria is common among the natives and may account for the large percentage of cases with splenic tumor.

The Wassermann reaction was tested in 91 cases and found strongly positive in 78 cases (or 85.7 per cent), moderately positive in 4 cases, weakly positive in 1 case and negative in 8 cases.

The 8 cases with negative Wassermann reaction were distributed with reference to diagnosis as follows: 2 cases were diagnosed B.; 1 case was diagnosed C+.; 1 case was diagnosed SL.: 1 case was diagnosed H.; 1 case was diagnosed H+.; 1 case was diagnosed G.; 1 case was diagnosed G+.

The Wassermann test was repeated in a few cases after one or two treatments. The interval between the two tests was not over a few weeks. In no case did the result of the second test differ from the first. If it had been possible to perform the test at a longer interval after treatment the results might have been different.

Thus it will be seen that in the small number of cases in which we have data on the Wassermann reaction the negative reactions all occurred in the late secondary stage, the tertiary stage, or in patients who had no active lesions but who gave a history of yaws.

Sequence and Duration of the Various Stages of Yaws. In the discussion of this entire series of cases we have tried to take up the various distinctive manifestations of the disease in the order of their development. In so chronic a disease as yaws it was obviously impossible, in the limited time at our disposal, to observe a single case from the appearance of the madre buba through the various stages to the final deformity produced by the gummatous lesions. Moreover it is apparent from the preceding sections that there is much overlapping of the various stages, many patients presenting two or even three different manifestations of the disease at the same time. In one case the madre buba persisted after the tertiary lesions had developed. Indeed not being able to observe the sequence of events we were often ignorant as to what

it had been. We believe, however, that a statistical study of the data collected enables us to unravel this riddle. Accordingly we have tabulated those cases that presented only a single manifestation of the disease, arranging them according to the average age of each group. The following table shows the extremes of age as well as the average and extremes of duration for each group:

Diagnosis.	No. of cases.	Age in years.		Duration.	
		Average.	Extremes.	Average.	Extremes.
M	15	6.6	$\frac{9}{12}$–12	2.3 mos.	1/12–7/12
B′	82	8.8	$1\frac{1}{3}$–50	12.3 "	1/12–7
B	60	9.3	1–42	29.6 "	4/12–17
C	327	21.6	3–68	9.0 yrs.	4/12–58
H	127	23.8	1–80	9.7 "	2/12–59
P	5	25.0	15–45	14.0 "	10–19
SL	37	28.8	5–80	13.5 "	5/12–39
G	67	29.3	3–85	16.3 "	7/12–59
	720				

This series comprises 720 cases and the number included in a majority of the groups is large enough to give averages that are probably fairly reliable. It seems to us that this arrangement according to average ages probably indicates the sequence in which the various lesions develop. Confirmatory evidence in support of the correctness of this view is furnished by the fact that the average duration of the disease at the time of observation in the various groups, with one exception (studded lesions), follows the same orderly progression as do the ages. The fact that the extremes of age and duration in each group may vary widely does not, in our opinion, invalidate the deduction which we have just made.

We have attempted to use the data contained in the above table to answer certain other questions. It will be noted that the average ages and average durations are given as of the date of observation, and in the case of duration this does not represent the actual average duration of the disease or any particular stage of it. To illustrate: A patient in the primary stage of the disease may have given a history of having had the madre buba two months at the time he applied for treatment; it is quite possible that this lesion would, without interference, have persisted for another two months. Thus in that particular case the duration of the primary lesion

would have been four months. If our reasoning is correct the actual average duration of the madre buba may be estimated by determining the average duration of the lesions at the time of observation in a sufficiently large number of unselected cases under natural conditions and multiplying this average by two. Thus in our series of madre cases the average duration at the time of observation was two and three-tenth months, and if 15 cases were a sufficiently large number for generalization we could say that the actual average duration of the madre stage of yaws is four and six-tenth months.

Furthermore if one stage supervened promptly on the termination of the preceding stage we could in turn calculate the actual average duration of each of the manifestations in the above series. For example we would arrive at the average duration of the B′ lesions by multiplying twelve and three-tenth months by two and subtracting the average duration of the madre stage. Thus we would find twenty months as the average duration of the florid secondary stage of the disease. The difficulty about applying such simple mathematical calculations to a disease like yaws is that there may be an interval free from active lesions and symptoms between the various stages of the disease on the one hand and a considerable overlapping of the stages on the other. This difficulty could have been overcome if it had been possible for us to obtain accurate histories of the date of onset of each stage of the disease. This we did not attempt, owing to the ignorance of a majority of the patients and the limited time at our disposal.

Although we obtained a history of the primary lesion persisting for six years in one case and saw patients with a florid secondary eruption in one case three years, in another seven years and in still another case nineteen years after the onset of the disease, if we may accept the history these patients gave, it seems probable that the average duration of the florid secondary eruption is less than two years and that the late secondary lesions, especially clavus, may recur throughout a long period which, while frequently much longer, averages about sixteen years, and that in some cases patients may live to extreme old age, eighty-five years in one case in our series, with tertiary lesions.

Result of Treatment. Neosalvarsan was used in the treatment of the cases here reported. The drug was dissolved in freshly distilled water in the proportion of 0.1 gram to 2 c. c. and injected within thirty to forty-five minutes after being put in solution. The intravenous method was used in all cases except young children with veins difficult of access. These cases received the injections intramuscularly in the buttock. The dose varied from 0.075 gram for an infant under one year of age to 0.6 gram for a fully developed adult. Intermediate doses were given in proportion to age and body weight. While reactions were frequent, including chill, fever, headache and general malaise, we observed no permanent ill effects, and in none of the cases which received the intramuscular injections did we observe abscess formation which reached the surface, although some of them showed a painful brawny swelling.

It should be remembered that we were working under field conditions with no facilities for hospitalizing patients even overnight, that many patients came two, three and even four days' journey on horse or on foot, over difficult trails, beat upon by tropical sun and often drenched by tropical rains, with the ground for a bed and the sky for a shelter. Oftentimes within an hour after receiving an injection these patients started on the return journey to their distant homes, again facing the hardships of the trail. The fact that no patient declined to return and receive further injections when advised to do so indicates as clearly as any statistics we might give either that the natives did not regard the reactions of a very serious nature or else that they considered the benefits to be derived outweighed the discomforts of the reactions.

From a statistical standpoint the results of treatment in our series is of little value, owing to the limited time over which the cases were observed and the fact that many cases received but a single treatment and were not seen again. The latter circumstance is easily explained. In the interior of Santo Domingo communication is largely by word-of-mouth and news spreads chiefly by means of the more or less chance wanderings of the natives. When we went up into the interior and set up our camp it was five days before the first patient, an old woman with "gomma," presented

herself for treatment. Two days later three children with a florid secondary eruption were brought to us. During the next few days patients ventured in in small numbers, but when the news of the results obtained in the early cases treated began to spread, patients came in rapidly increasing numbers. By the end of the first month we were getting an average of about 25 to 30 new cases a day and during the last two weeks of our stay the daily number of new and old cases ranged between 100 and 300. In spite of the fact that we sent out notices well in advance of our departure, to stop new cases from coming in, the daily number increased almost up to the last day of our stay. Their appeal for help was so piteous and their condition so distressing that we could not refuse them a single injection, although we knew that we would never see them again. This accounts for the large number of cases in which we have no note as to the result of treatment.

It has been claimed that 90 per cent of yaws cases can be cured by a single injection of salvarsan. Our experience was not so favorable as this, though if we had had a longer time in which to observe the results we could have allowed a longer interval between injections and might have found it necessary to repeat the treatment in a smaller percentage of the cases. Moreover the number of patients requiring more than one injection to effect a cure might have been fewer if 0.9 gram neosalvarsan instead of 0.6 gram had been adopted as the adult dose.

We have designated our results as cured, practically cured, much improved, improved and unimproved. No case was recorded as cured unless all the lesions (scars excepted) had entirely disappeared. If there remained at our last opportunity of observation only so much as a few black crusts almost ready to fall, where a week or two before there had been an abundant crop of fresh granulomata, we designated such cases as "practically cured," although we were convinced that could we have seen these cases a week later, without further treatment, they would have fallen in the cured list. The remaining designations—much improved, improved and unimproved—are sufficiently clear without further comment except to say that due conservatism was used in applying them, and that

if error was made it was probably on the side of underrating the actual improvement.

As to the permanency of the results obtained we have no observations, and even if the present condition of these patients could be ascertained sufficient time has not elapsed to determine the permanency of the cures. We believe, however, that Ehrlich's ideal of "sterilizans magnum" is more nearly obtained with salvarsan or neosalvarsan in yaws than in the treatment of any other disease with any other drug.

There are obvious difficulties in giving a statistical statement of the results of treatment which can be correctly interpreted. As previously stated many of the cases which were designated as "practically cured" might have fallen in the "cured" column without further treatment if there had been an opportunity for examining them after the lapse of another week. Moreover the percentage of cures after one injection might have been materially increased if we had allowed a longer interval of time in which to determine the result of the first injection.

The interval between injections was one week in the majority of cases, exceptionally it was as short as five days and in a moderate number of cases it was ten days or two weeks. The final result of treatment, as far as we had opportunity to observe it, was usually made one week after the last injection.

The following table gives the results of treatment in the various groups as noted at the last observation on each case and shows the number of injections each patient had received at the time of this note. The 127 cases which gave a history of having had yaws, but who had no active lesions of the disease at the time they presented themselves for treatment, are not included in this table, as we had no means of judging of the results of treatment in these cases. We have also excluded from the table 31 miscellaneous cases of yaws in which the results of treatment were difficult to determine, owing to complicating diseases or for other reasons. Exclusive of these two groups there are 888 cases, and we have notes as to the result of treatment in 570 of these.

TABLE SHOWING RESULT OF TREATMENT IN 570 CASES.

Diagnosis	No. of cases.	Results.		Cured.				Practically cured.				Much improved.				Improved.				Unimproved.			
		No note.	Noted.	No. of doses.				No. of doses.				No. of doses.				No. of doses.				No. of doses.			
				1	2	3	4	1	2	3	4	1	2	3	4	1	2	3	4	1	2	3	4
M	15	4	11	2	..	..	..	5	2	1	..	..	1										
MB′	25	10	15	2	2	..	..	4	1	..	..	6											
MB′C	13	2	11	..	3	..	..	2	3	..	..	2	..	..	..	1							
MB	6	3	3	..	..	..	..	..	..	..	..	1	..	1	..	1							
MBC	5	3	2	..	1	..	..	..	..	..	..	..	..	..	..	..	1						
B′	82	16	66	13	11	1	..	14	6	1	..	8	3	2	..	5	1	..	..	1			
B′C	24	4	20	2	4	..	..	5	5	..	..	3	1										
B	60	24	36	4	9	2	..	1	3	..	..	11	3	..	..	3							
BC	106	26	80	..	10	1	..	7	6	..	..	36	7	1	..	7	3	1	..	1			
BCP	13	7	6	..	3	..	..	..	..	..	..	3											
C	327	143	184	5	5	5	1	3	9	..	..	27	16	..	..	55	10	..	..	41	7		
C+	41	13	28	..	1	1	..	1	..	..	..	7	3	3	..	4	2	..	..	6			
CP	50	15	35	..	..	1	..	..	1	1	..	5	7	..	..	10	1	..	..	9			
P	5	3	2	..	..	..	..	..	..	..	..	1	..	..	..	1							
S.L	37	17	20	1	2	1	..	1	..	1	..	3	2	..	..	4	2	..	..	3			
G	67	24	43	..	..	..	..	..	..	..	..	..	4	5	2	8	..	5	..	15	3	1	
G+	12	4	8	..	..	..	..	..	..	..	..	..	4	1	..	..	1	..	..	2			
	888	318	570	29	51	12	1	43	36	4	..	113	51	13	2	99	21	6	..	78	10	1	
				93				83				179				126				89			

Briefly summarized, irrespective of the number of injections each patient received, this table shows the following results:

	Cases.	Per cent.
Cured	93	16.31
Practically cured	83	14.56
Much improved	179	31.40
Improved	126	22.10
Unimproved	89	15.61
Total	570	99.98

There were 362 cases in the above series in which the final result noted was after a single injection of neosalvarsan. Briefly summarized they are shown in the following table:

RESULTS AFTER A SINGLE INJECTION OF NEOSALVARSAN.

	Cases.	Per cent.
Cured	29	8.01
Practically cured	43	11.88
Much improved	113	31.21
Improved	99	27.34
Unimproved	78	21.54
Total	362	99.98

Perhaps one gets a better idea of the efficacy of treatment in yaws by considering the cases which showed that no improvement followed the administration of neosalvarsan. In the above series of 570 cases only 1 patient (a gomma case) showed no improvement after three injections and only 10 patients (3 gomma and 7 clavus cases) showed no improvement after two injections. This is certainly in marked contrast to the 113 cases which showed marked improvement after but a single injection.

We have further analyzed our figures to see if they will show what stage of the disease is most readily amenable to treatment. For this purpose it seems probable that more reliable information will be obtained by combining the cured and practically cured groups and considering only those cases in which the result noted was obtained after a single injection. They are summarized in the following table:

Diagnosis.	Total No. cases.	Average age, years.	No. of cases, results noted.	Cured, or practically cured, after one injection.	
				Number.	Per cent.
M	15	6.6	11	7	63.63
MB′	25	8.5	15	6	40.00
MB′C	13	11.6	11	2	18.18
MB	6	24.2	3	0	
MBC	5	10.0	2	0	
B′	82	8.8	66	27	40.91
B′C	24	12.9	20	7	35.00
B	60	9.3	36	5	13.88
BC	106	16.5	80	7	8.75
BCP	13	20.5	6	0	
C	327	21.6	184	8	4.34
C+	41	29.4	28	1	3.57
CP	50	32.7	35	0	
P	5	25.0	2	0	
SL	37	28.8	20	2	10.00
G	67	29.3	43	0	
G+	12	25.2	8	0	

The number of cases comprising some of the groups in the above table is too small to be of value for statistical purposes, but, taken as a whole, the figures seem to show what we would expect, *i. e.*, the earlier the stage the more readily does it respond to treatment.

DISCUSSION

Dr. E. J. Wood, Wilmington, N. C.: Yaws was described by John Brickell, M.D., in his *Natural History of North Carolina*, published in 1737. The disease has recently been recognized in the same region. It has brought up the question whether or not a portion of the very large Wassermann positives in negroes of the Southern States may not be due to yaws, which gives fully as positive response as does syphilis.

Dr. David Riesman, Philadelphia: We have had one case in the Philadelphia General Hospital—a white soldier who had picked the disease up in France. His blood gave a strongly positive Wassermann reaction. Dr. Moss did not refer to this feature.

Dr. S. Solis-Cohen, Philadelphia: I should like to ask Dr. Moss about the treatment.

Dr. Joseph Collins, New York: Has Dr. Moss had opportunity to investigate any of the terminal cases from the pathologic standpoint to determine if the lesions are of the peripheral nerves or the muscle substance and not of the spinal cord? That would make an important differentiation from syphilis, because syphilis very rarely produces the former lesions.

PLAGUE-LIKE INFECTIONS IN RODENTS

SUMMARIZED REPORT

By W. T. COUNCILMAN, M.D.
AND
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Plague has occurred during the past year in Galveston, Beaumont, New Orleans and Pensacola, as well as in a number of Mexican and Central and South American ports.

At a special conference of federal, state and municipal health authorities held in August, 1920, relative to the plague situation in the United States, it was resolved in part that a nation-wide effort should be made at the present time to determine as accurately as possible the presence or absence of rodent plague in all ports of entry and principal centers of railway freight transportation in the United States, and, further, in order that such investigative work and the concomitant preventive measures against the spread of rodent plague shall be carried out in a concertedly effective manner, the Surgeon-General of the United States Public Health Service be requested to send out to all state governments suggestions as to the most efficient eliminative method of carrying on rat surveys and the practical manner of permanent plague prevention. It was also resolved that the present serious, even though not alarming, bubonic plague situation in the southern states should be the occasion for inaugurating a nation-wide campaign against rodents on economic as well as public health grounds.

Our Public Health Service in November, 1920,[2] also called attention to the fact that with bubonic plague, both human and rodent, actually known to be present in several parts of the United States, it would seem to be both wise and expedient for cities and towns throughout the country to determine the disease status of their

[1] Illustrated by lantern slides.

[2] Public Health Reports.

rat population. As an arbitrary standard of trapping, it has been suggested that 10 per cent of the estimated rat population be fixed as the number to be examined, and that the examination of 1000 rats per 10,000 human population will afford reliable evidence of the occurrence of plague.

If these investigations are carefully carried out, diseases other than plague are likely to be occasionally encountered in these rodents, and it is particularly on account of the importance which these rat surveys have assumed in the United States that our investigations of the diseases which more or less resemble plague in rodents have been undertaken. Therefore these studies have a practical significance.

A number of different spontaneous infections in rats and other rodents have been reported from time to time, in some of which the organisms isolated have considerably resembled B. pestis. In many of these the causative organisms have shown bipolar staining, have been Gram-negative and similar in their cultural appearances to B. pestis.

The microörganisms giving rise to infection in rodents which may simulate considerably plague infection may be classified as follows:

ORGANISMS CAUSING PLAGUE-LIKE INFECTIONS IN RODENTS

I. The hemorrhagic septicemia group (Pasteurella group):
   1. Bacillus of chicken cholera.
   2. Bacillus of rabbit septicemia.
   3. Bacillus of swine plague (contagious pneumonia).

II. Bacillus of pseudotuberculosis rodentium.

III. The Gaertner-paratyphus group (Salmonella group):
   1. B. enteritidis Gaertner.
   2. B. paratyphosus β.
   3. Bacillus of hog cholera (B. enteritidis Aertrycke).

   Bacillus of Danysz, B. typhi murium, B. psittacosis, B. icteroides — Identical or mixtures of the above organisms.

IV. B. mucosus capsulatus (Friedlaender's bacillus).

V. B. tularense.

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During the time at our disposal today we can bring to your attention but one of these plague-like infections, namely, that caused by B. tularense. McCoy and Chapin, in 1911 and 1912, when searching for plague in rodents, discovered and described a disease found in the California ground squirrels which is experimentally transmissible to other rodents and which in practically all susceptible animals gives rise to an infection which presents lesions very similar to those produced by B. pestis. Subsequently they isolated an organism from this infection which they called B. tularense, and which they finally cultivated upon egg-yolk but not on other media. However, comparatively little attention has since been paid to this disease and to the very important work of McCoy and Chapin on the subject. There are in the literature only four other reports besides those of Chapin and McCoy of investigations relating to infection with this organism. Of these four publications, two are by Wherry and Lamb, and one by Vail, in 1914, relating to two cases of severe human conjunctivitis in which B. tularense was apparently the infecting organism. In the one human case reported in detail by Wherry and Lamb and by Vail, B. tularense was not directly isolated from the patient, but was subsequently found in some of the later animals of a series of which the first guinea-pig was inoculated with scrapings from the conjunctival abscess of the left eye. The submaxillary and cervical glands of the same side of the neck of this patient became swollen and a septic temperature varying between 100° and 102.6° developed. The temperature apparently lasted for eight to ten days. The patient left the hospital and subsequently recovered.

The fourth and most recent of these publications regarding this organism has been made by Dr. Edward Francis,[1] of the Public Health Service, in connection with a disease of man known as deer-fly fever.[2] Dr. Francis writes as follows: "In recent years there has occurred among the rural population of Millard County, Utah, a disease initiated (according to popular belief) by a fly bite on some

[1] Public Health Report, September, 1919.

[2] Dr. Brues informs me that the small biting house fly with spotted wings, genus Chrysops (Brues) is perhaps referred to under this designation.

exposed surface of the body, manifested by the enlargement of the lymph glands which drain the bitten area, and by a fever of a septic type lasting from three to six weeks. The site of the bite and the affected lymph glands become tender and inflamed, and they commonly suppurate. There is marked prostration and the patient is confined to his bed. Probably two dozen cases occurred in Millard County in each of the years 1917, 1918 and 1919. The first case known to have terminated fatally was reported in 1919." Dr. Francis selected a typical case of deer-fly fever, and from this case two series of guinea-pigs and rabbits were inoculated. The first series was inoculated with the patient's blood and the second series with pus obtained from a suppurating cervical gland of the patient. Both series of animals developed a disease which proved fatal in a few days. At postmortem these cases exhibited caseation of the lymph glands and small necrotic foci throughout the liver and spleen. Subinoculations from the infected animals into healthy ones invariably reproduced the same lesions. Dr. Francis isolated from these animals an organism which grew upon egg-yolk and which he regarded as probably B. tularense. No description is given in this preliminary report of an attempt to cultivate this organism directly from patients suffering with deer-fly fever.

Today we are particularly concerned with infection of rodents with this organism, regarding the pathological histology of which infection apparently no detailed publication has been made previously. The British Indian Plague Commission laid stress upon making the diagnosis of plague infection in rats with the naked eye, and emphasize particularly the following changes:

1. Presence of rigor mortis, subcutaneous congestion, hemorrhages and edema.
2. Changes in the lymphatic glands (buboes).
3. Characteristic appearances in the abdominal organs.
4. Characteristic appearances in the organs in the thorax.

In regard to the changes in the lymphatic glands in the acute type a bubo was found in 85 per cent. The presence of the bubo is the most important sign of plague in rats.

The next most important sign is the condition described as the

PLATE II

Fig. 1 Bacillus tularense from a culture.

Fig. 2. Cultures of Bacillus tularense 5 and 10 days after inoculation of the media.

Fig. 3. Spleen in Bacillus tularense infection.

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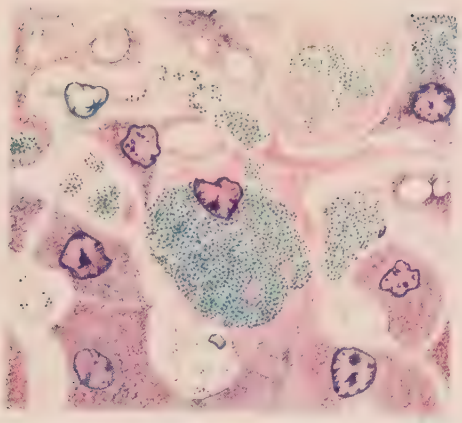
granular liver, the appearance of which is due to small necrotic foci scattered through its substance. The gray or whitish granules are most easily observed on the surface and the organ appears as though dusted with gray pepper. More or less fatty degeneration is also usually present. The enlargement of the spleen is usually marked.

The changes in the organs of the thorax to be noted are the presence of hemorrhages in the lungs and heart and the appearance of pleural effusion. Owing to the fact that in the naturally infected plague rats the bacilli are often not present in large numbers and their distribution is not constant, the Indian Plague Commission believed that for the purpose of diagnosis a naked-eye examination by a competent observer is more satisfactory than a microscopic one alone. We will demonstrate by lantern slides that it is generally practically impossible to differentiate B. tularense infection from infection with B. pestis in certain rodents by the naked eye alone. This picture (Plate I) illustrates a guinea-pig which was inoculated by lightly scarifying the skin of the abdomen with a scalpel and rubbing in, as in vaccination, through the abrasions in the skin a small portion of the spleen of another guinea-pig which had succumbed to tularense infection. We have had a wide experience with plague infection in rodents, and it is frequently impossible to tell at autopsy, from the macroscopic lesions, in many instances whether B. tularense or B. pestis was the infecting organism. Also, as the bacilli in plague infection of rats are often not present in large numbers, or postmortem bacilli may have invaded the tissues, additional difficulties in diagnosis may thus be encountered. Stitt points out that the crucial test for any plague material is the power of the plague bacillus to infect a rat or guinea-pig when the material is rubbed into the shaven skin of the animal. This is true for rats when the infection is exceedingly virulent, but such an inoculation in a guinea-pig would not differentiate tularense infection from plague.

When a guinea-pig is inoculated by scarifying the skin of the abdomen with a scalpel and rubbing the scarified area with a small portion of the spleen of an infected animal, death usually occurs on the third or fourth day after the inoculation, but the day of

death may vary between three to six days after inoculation. A series of thirty-seven guinea-pigs has been successively inoculated in this manner and all of the animals have died of the disease. At the autopsy there is induration and thickening about the point of inoculation, subcutaneous congestion, edema and often hemorrhages of varying amount. The adjacent inguinal lymphatic glands are swollen, and frequently there are hemorrhages in the region of the glands. The glands are often softened and necrotic, and upon section the center is yellow and caseous in appearance. The spleen (Plate II, Fig. 3) is greatly enlarged and congested, and both it and the liver (Plate III), which is also congested, contain very numerous necrotic foci which vary from pin-point to about 1 mm. in diameter. The lungs and also the kidneys frequently show hemorrhages. Very large numbers of a very minute round or rod-shaped organism are found in microscopic preparations from the heart's blood and the liver, spleen and swollen lymphatic glands. (Plate II, Fig. 1). These organisms stain with carbol fuchsin and anilin-gentian violet and with Giemsa's solution. Prolonged staining gives the best results. They are decolorized by Gram's stain and are apparently not motile. In stained preparations the organisms are found lying inside well-defined, round or oval, clear, unstained areas; sometimes several or a dozen or more organisms are found in such a clear space. In size the rod-shaped organisms vary from about 0.3 to 0.7 μ in length and 0.2 to 0.3 μ in width. The round forms vary from 0.2 μ to 0.5 μ in diameter. We have been successful in cultivating this organism on coagulated egg-yolk or more rarely on a medium made with the liver and spleen of normal guinea-pigs, and nutrient beef agar. This medium is made by grinding up the liver and spleen of a normal guinea-pig in a mortar with sand, adding normal saline solution, filtering through a Berkefeld filter and then adding equal parts of 3 per cent nutrient beef agar at a temperature of about 55° C. The organisms grow very slowly, colonies appearing after three or four days in the incubator at 37° C. These colonies become slightly larger and frequently coalesce after an interval of a week to about ten days. The colonies which at first can often only be seen by a hand lens may eventually attain a diameter of 1 to 2 mm. They appear very moist, translucent and drop-like in character. (Plate II, Fig. 2.)

PLATE III



Section of liver and infection with Bacillus tularense.

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The histology has been studied by infecting animals with B. tularense and killing them after one, two, three or four days and examining the tissues of these and also animals that have died naturally. Very briefly the sections show necrosis and polymorphonuclear infiltration in the vicinity of the site of the inoculation, followed by an extensive proliferation of the vascular endothelium.

In the lymph nodes, liver and spleen there are miliary foci formed by accumulation of mononuclear cells followed by necrosis and infiltration with polynuclears. In the liver there are in addition foci of necrosis without cell accumulations of capillary lesions. These changes are constant, and there may be lesions of much the same character in the adrenal, the heart, the lungs and the testicle. After death, B. tularense is present in great numbers. There is a general infection of the bloodvessel endothelium and the organisms may be found in vessels of any part of the body. In addition the organisms pass from the endothelium into the cells of the liver, which they gradually destroy and replace, forming large globular masses of bacilli which are easily demonstrable and may be seen even with the objective AA as deeply staining areas. They form the most striking histological feature of the disease. (Plate III.) A similar process takes place in the adrenal glands, the cortical cells being chiefly involved. The glomeruli of the kidney are also infected and accumulations of mononuclear cells are found in them, the lesion being not unlike that of intracapillary glomerular nephropathy in men. The essential lesion is infection of the endothelial cells, general but more marked in the vessels of certain organs. Polymorphonuclear leukocytes play but a secondary role, and very rarely contain bacilli. The staining of bacilli in the tissues is difficult, and prolonged staining with Giemsa's solution gives the best results. Owing to the limited time the more detailed description of the histological appearances cannot be presented today and these will be published in the completed report.

We wish to express our appreciation to Dr. G. W. McCoy, Director of the Hygienic Laboratory, U. S. Public Health Service, Washington, who has on several occasions furnished us with a strain of B. tularence. [The gross and microscopic lesions were fully illustrated by lantern slides.]

DISCUSSION

Dr. W. W. Ford, Baltimore, Md.: We recently had a colored man come in to the venereal disease clinic complaining of severe fever, and on examination found that his temperature was 104°. There was no evidence of venereal infection, but he gave an obscure history of syphilis ten to twelve years previously. There is very marked enlargement of the inguinal and axillary glands. The picture was suggestive of bubonic plague. The material aspirated from the glands gave organisms with cultural reactions which seemed to be those of Bacillus pestis. We were puzzled what to do. The man had been a dock laborer and presumably had been exposed to rat bites. Dr. McCoy decided that the organism was not plague on the basis of the lack of virulence for animals. We thought it was a non-virulent plague organism. In cases of this character what should one do? We must establish a diagnosis at the time the case occurs. From the standpoint of city administration we should hesitate to report it as plague, because of quarantine regulations. The case was observed for a long time and the man recovered spontaneously. He did not have plague. This brings up difficult questions in regard to plague. In Brazil they have plague-like organisms which cause slight inflammation. Inflammation is not necessarily diagnostic. We are at the beginning of our study of plague and plague-like organism in this country.

Dr. H. Noguchi, New York City: Was this a direct-contact infection? This organism is one-fifth of a micron in size. Will it assume a form which will pass through an ordinary filter?

Dr. William H. Park, New York: In New York City itself we have not had a case of plague in thirty years, but there have been a few cases on vessels in the harbor. Every few years we examine some thousands of rats, and this costs money and labor. In summertime some of the rats are partially decomposed when delivered at the laboratory, and it is a disgusting task to separate them from those fit to examine. We have never found a rat infected with the plague bacillus. I think if the health authorities of coast cities would examine from time to time a thousand rats enough has been attempted. Rats apparently must be badly infected with plague before human beings are apt to contract it. I fear that the amount of examination suggested would take the public health worker's time from more important things. The health department of New York City has seven rat catchers at work. If no infected rats are found within three months we will stop the rat examinations.

Dr. R. P. Strong: In reply to the question regarding the method of infection in deer-fly fever, this has not yet been conclusively demonstrated. It has been supposed that the infection was transmitted by some biting insect, to which fact I have already referred. Bacillus tularense will not pass through porcelain filters and filtrates of cultures of this organism are not infective for guinea-pigs. McCoy and Chapin first demonstrated this fact.

In reply to Dr. Park's question, I think that unless there is absolutely no doubt of the nature of the organism in a suspected case of plague occurring in this country that the cultures should be immediately sent to the Public Health Laboratory at Washington for examination before a diagnosis of the case is announced.

In regard to the method recommended for the detection of rodent plague in a community referred to in my remarks, I should say that I was not present at the meeting of the health officials held at Galveston and Beaumont, Texas, which made these recommendations.

I agree with Dr. Park that a few bacteriologic examinations carefully performed would be of greater value than the incomplete examination of thousands of rats, many of which are badly decomposed.

EXPERIMENTAL PIGMENT CIRRHOSIS DUE TO COPPER AND ITS RELATION TO HEMOCHROMATOSIS[1]

By F. B. MALLORY, M.D., FREDERIC PARKER, Jr., M.D.
AND
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Synopsis:

Introduction.
The primary cell lesion in hemochromatosis.
Pathogenesis.
Technical methods employed.
Previous attempts to produce hemochromatosis.
Experimental pigment cirrhosis due to chronic poisoning with copper.
Discussion.
Summary.
Conclusions.
Bibliography.

Introduction. Hemochromatosis is a chronic disease characterized by the presence of two pigments in the cells of various organs and tissues of the body. One of the pigments, hemosiderin, gives the reactions for iron; the other, hemofuscin, does not. The accumulation of these pigments in certain cells leads eventually to three characteristic clinical signs: cirrhosis of the liver, sclerosis of the pancreas with resulting diabetes mellitus, and pigmentation of the skin. It is on the simultaneous presence of these three features in a typical case that a positive diagnosis of the disease is based during the life of a patient.

Well-marked cases of hemochromatosis are rare—less than a hundred are on record in the literature at the present time. On

[1] From the Pathological Laboratory of the Boston City Hospital. The expenses of the work presented here have been paid in part from a research fund received from Dr. and Mrs. Henry F. Sears.

the other hand the milder or earlier type of the condition, recognized by the pathologist as hepatic hemosiderosis, or pigment cirrhosis, in which the deposition of pigment is limited entirely or chiefly to the liver, is much more common than is generally appreciated, but there is nothing to call attention to it clinically.

The following statistics from the pathological laboratory of the Boston City Hospital are of interest in this connection:

Total number of autopsies during the past twenty-five years	4507
Total number of cases of well-marked cirrhosis of the liver	224 (5.08 per cent)
They are classified as follows:	
Alcoholic cirrhosis (hyalin)	104 (46.42 per cent)
Pigment cirrhosis (hemosiderin)	16 (7.14 per cent)
Alcoholic and pigment cirrhosis combined (hyalin and hemosiderin)	50 (22.32 per cent)
Syphilis	14 (6.25 per cent)
Infectious (colon bacillus)	10 (4.46 per cent)
Acute yellow atrophy type	8 (3.57 per cent)

In this series fourteen cases of central (sometimes called cardiac) sclerosis and one of chronic perihepatitis are not included.

These additional statistics, showing the further occurrence of alcoholic hyalin and of hemosiderin, are of value.

	Slight cirrhosis.	No cirrhosis.
Alcoholic hyalin	44	10
Hemosiderin	8	1
Alcoholic hyalin and hemosiderin	13	6

In other words the hyalin characteristic of so-called alcoholic cirrhosis was present alone or associated with hemosiderin in 227 of the 4507 livers, or 5.14 per cent; while hemosiderin was found likewise alone or associated with alcoholic hyalin in 94 livers, or 2.08 per cent.

These additional cases listed above represent the early stages of lesions which if continued would eventually have terminated in typical examples of cirrhosis.

One point must be borne in mind in connection with these statistics: Of the 4507 autopsies 1057 were made on children from the infectious wards (scarlet fever, diphtheria, etc.). If they are excluded the percentages of livers showing the hyalin and hemo-

siderin which are characteristic of alcoholic and pigment cirrhosis are considerably larger, namely, 6.57 per cent and 2.72 per cent.

The amount of alcoholic hyalin and of hemosiderin present in the livers classified here varied greatly from a slight amount up to an extreme degree. Of the cases of pigment cirrhosis four showed very general distribution of the hemosiderin, so that they deserve to be included under the term hemochromatosis. One case was pure in type, two showed only a very slight amount of alcoholic hyalin and the fourth a marked degree of it.

Hemochromatosis has excited much scientific curiosity ever since the condition was first recognized. The earliest cases on record were reported by Trousseau early in the eighteenth century, Troisier in 1871, Hanot and Chauffard in 1882, and Hanot and Schachmann in 1886. The name and the first adequate description of the disease are due to von Recklinghausen in 1889, who noted the presence of the two different kinds of pigments and devised the word hemofuscin for the one which does not give the iron reaction. The French had already employed the term diabète bronzé (bronzed diabetes) for the same condition, and it is still in common use.

The best-known papers on the subject in English are by Maude E. Abbott, Opie and Sprunt. They consist, like most of the others, of case reports with gross and microscopic descriptions of the lesions found postmortem, references to the literature and discussions of the pathogenesis of the condition.

The Primary Cell Lesion in Hemochromatosis. In order to attempt to produce hemochromatosis experimentally in animals it is obviously important to have, if possible, a perfectly clear understanding of the nature of the lesion as it occurs in man.

The study of a considerable series of cases shows definitely that the process of pigmentation makes its appearance first in the liver; then much later it appears more slowly and in varying degrees of intensity, but more or less synchronously, in the pancreas, heart, kidney, adrenal glands, mesenteric lymph nodes, gastrointestinal tract, skin and other organs and tissues. The lesion develops so slowly that it is difficult to follow every detail of the changes produced, but enough is evident for a full comprehension of the process. The liver is the best organ in which to study the lesion because,

owing to active and recurrent regeneration of the liver cells, all stages of the effect produced by the deposition of the pigment can be followed.

(*a*) *Liver.* In the liver the pigment is deposited first in the endothelial cells lining the sinusoids and in the liver cells, then in the epithelial cells of the smallest bile ducts and in some of the fibroblasts of the stroma. Later and much more slowly it appears in the smooth muscle cells of the bloodvessels, in the fibroblasts around the larger vessels and ducts and finally in the epithelial cells lining the large bile ducts.

The pigment granules are located at first in the cytoplasm of the liver cells close to the bile capillaries, but later may fill the whole cell. One point is quite noticeable: in the early cases the pigment is deposited uniformly throughout the liver in all the liver cells, but more abundantly, as a rule, at the peripheries of the lobules than in the centers.

After the accumulation of pigment has reached a certain degree of intensity in the liver cells they begin to undergo necrosis singly here and there or in small groups. The necrotic cytoplasm is dissolved by the action of leukocytes and then the pigment granules are taken up in great numbers by endothelial leukocytes which tend to migrate with their contents to the periportal connective tissue, where they accumulate in the lymph vessels and between the connective-tissue cells of the stroma and attempt to digest the pigment.

The number of endothelial leukocytes filled with pigment which are present in the stroma varies greatly in the different cases. Sometimes they are few in number, at other times very abundant. The number seems to depend on the severity and duration of the process. In early active progressing lesions they are numerous. In cases of long duration where the process has been slight or intermittent the endothelial leukocytes have had time to digest and remove the hemosiderin they contained and to disappear themselves.

As the liver cells die off regeneration takes place, often in the form of islands of new cells which may attain a size of 1 or 2 mm. or even more. These new cells at first are not pigmented. Then granules of light yellow pigment appear which do not give the iron reaction but stain more or less intensely with basic anilin dyes, especially

with methylene blue, fuchsin and safranin. At a later stage the granules gradually change so that they no longer stain with the anilin dyes but give the iron reactions with ammonium sulphide and with ferrocyanide of potassium and hydrochloric acid. It is often possible to find in the comparatively young regenerated liver cells the two kinds of pigment granules in adjoining cells or mixed in various proportions in the same cells. The same is true of the liver cells in early cases of hemochromatosis. In other words the regenerating foci of liver cells show definitely that hemofuscin is deposited first and is then gradually transformed by the metabolic activity of the cells, so that the hemoglobin compound is broken down and hemosiderin is formed, rendering possible the iron reaction. The presence of the two kinds of pigment granules in the liver cells of early cases of hemochromatosis strengthens this interpretation.

It is this primary pigment, hemofuscin, which is deposited much more slowly and hence relatively much later in the smooth muscle cells of the bloodvessels, in the fibroblasts around the large vessels and ducts and in the epithelial cells lining the large bile ducts. All these cells reduce the hemofuscin with great difficulty, so that hemosiderin is formed only here and there. A possible explanation is the relatively greater distance of all these cells from the active circulation; their metabolic activity is probably less.

One other thing can be learned from the lesion in the liver. The islands of regenerating liver cells have one point in common with tumors; as they grow they are able to obtain by proliferation from the old stroma a new stroma of connective tissue and sinusoids of their own. In this way the amount of connective tissue in the liver is gradually increased. At the same time as the old pigmented liver cells die off their stroma is left behind to shrink and coalesce. In this way the apparent increase of connective tissue in foci, that is, the scleroses, in hemochromatosis (and also in the alcoholic type of cirrhosis) are formed, not because of any direct proliferation of fibroblasts to take the place of the liver cells which have disappeared, as is so generally taught.

There is no evidence of necrosis of fibroblasts owing to the deposition of pigment in them or of regeneration of them in consequence.

(b) Pancreas. The lesion in the pancreas is much like that in the liver. Hemofuscin is deposited fairly readily in the cells of the glands and of the islands, but much later in the epithelium lining the ducts. It is easily changed to hemosiderin and leads eventually to necrosis of the cells followed by regeneration. In one case mitosis of island cells was of fairly common occurrence and was present also occasionally in the gland epithelium. The pigment from the necrotic cells is taken up by endothelial leukocytes which migrate with it to the stroma, especially around the larger blood-vessels, where they accumulate in considerable numbers. As the gland cells are killed off the stroma which is left behind shrinks and condenses, so that foci of compact connective tissue, scleroses, are formed. Hemofuscin is found in slight amount, chiefly in fibroblasts in the stroma.

As is well recognized the pigmentation and sclerosis in the pancreas lead eventually, if the patient lives long enough, to diabetes mellitus. This outcome is of much scientific importance, because hemochromatosis is the one definite process which produces this condition. It is to be noted that the glands are much more affected than the islands.

No evidence of fibrosis or of disappearance of the islands can be found. In advanced cases of hemochromatosis, however, when diabetes has occurred some of the islands present the typical hyaline transformation. This lesion in the islands suggests strongly that the hyaline change is the result and not the cause of the diabetes.

(c) Heart. The pigment deposit takes place in the endothelial cells of the capillaries and in the muscle fibers. In the latter it is most abundant at first in the cytoplasm at the ends of the nuclei, but later appears distributed often very uniformly between the striations throughout many of the fibers. When the pigmentation has reached a certain degree of intensity the portion of a fiber most involved undergoes necrosis and the pigment granules are taken up by endothelial leukocytes which, after a time, migrate with them to the stroma and there remain. No regeneration of muscle takes place. In time the stroma shrinks, the capillaries disappear to some extent at least and foci of sclerosis, which may be fairly extensive, are formed by the contraction of the old stroma.

It is doubtful if the lesion, which is always a late one, causes any clinical symptoms. The scleroses are like those occurring as the result of impaired nutrition due to sclerosis of the coronary vessels, generally included under the term chronic myocarditis.

(*d*) *Adrenal Glands.* The pigment is deposited almost exclusively in the epithelial cells in the outer layer of the cortex, that is, in the zona glomerulosa. Whether or not actual necrosis of the cells is produced cannot be determined.

(*e*) *Kidney.* The pigment granules are found chiefly in the epithelial cells lining the distal convoluted tubule and the ascending limb of Henle's loop of each renal element. They also occur in a few of the endothelial cells lining the capillaries in the glomerular tuft.

The lesion in the kidney is never marked because the renal cells destroyed by the presence or action of the pigment desquamate and are replaced by regeneration. There is no destruction of tubules. The pigment escapes with the urine and does not have to be taken care of by endothelial leukocytes.

(*f*) *Retroperitoneal Lymph Nodes.* The pigment is brought to the lymph nodes chiefly through the lymphatics, which drain from the liver and pancreas. It is carried in endothelial leukocytes which have obtained it through necrosis of hepatic and pancreatic cells. These leukocytes pass through the peripheral and other lymph sinuses and invade the lymphoid tissue, where they often accumulate in great numbers, causing, apparently mechanically disappearance of the lymphoid cells and increase of connective tissue in the same way as is done by carbon carried to the peribronchial lymph nodes.

(*g*) *Skin.* Hemosiderin occurs in fibroblasts around the coil glands and occasionally elsewhere, in the fat cells of the corium and rarely in the epithelial cells of the coil glands, but not in the cells of the epidermis. Its presence in the skin is of much clinical importance because it renders possible a positive diagnosis during life. The chemical tests for iron can easily be made on sections of an excised bit of tissue, preferably from a well-pigmented site.

(*h*) The pigment in hemochromatosis is deposited in various other organs and tissues besides in those already described, but

they can be summarized briefly under the different types of cells involved.

In severe cases pigment is present, chiefly in the form of hemosiderin, in both types of epithelial cells of the stomach, in the thyroid and parathyroid and in the ependymal cells of the choroid plexus.

Fibroblasts quite commonly contain pigment; in some situations, as around the coil glands, the granules give the iron reaction; in other locations, as in the capsules of organs and around large bloodvessels, hemofuscin is more often present.

The smooth muscle cells of bloodvessels and of the gastrointestinal tract often contain much pigment; most of it is hemofuscin, but in places the granules give a good iron reaction.

The skeletal striated muscle fibers sometimes contain pigment usually located close to the nuclei which often undergo direct division and multiply considerably. Rarely necrosis of fibers occurs and endothelial leukocytes take up the pigment.

Fat cells very generally everywhere, but especially perhaps in the subcutaneous tissues, contain pigment which is present in the form of hemosiderin.

Endothelial cells lining capillaries are the first almost everywhere to take up the pigment. It is always in the form of hemosiderin because the hemofuscin in these cells is so quickly broken down. The pigment appears first in the endothelial cells of the liver, later in those of the heart, spleen, bone-marrow and other organs.

The endothelial leukocytes obtain their pigment almost entirely from the destruction of the epithelial and other cells which were filled with it. Some of them, however, may arise from desquamated endothelial cells filled with the granules.

Hemosiderin also occurs in the ganglion cells of the central and sympathetic nervous systems and in combination with hemofuscin in the peripheral nerves.

Interpretation. The primary essential lesion of hemochromatosis is the deposition of a pigment of hemoglobin origin in various cells of the body beginning with those in the liver. The pigment is hemofuscin; it gives no iron reaction. In many kinds of cells (hepatic, pancreatic, endothelial, etc.) it is quickly broken down so

that from it is formed hemosiderin which gives the iron reaction. In certain cells (fibroblasts in various locations, smooth muscle cells, epithelial cells lining the larger bile ducts) the primary pigment is broken down slowly or not at all and persists more or less indefinitely as hemofuscin.

When the deposit of pigment reaches a high degree it causes necrosis of certain parenchymatous cells (liver, pancreas, heart); the necrotic cells are invaded by endothelial leukocytes which take up the pigment in great quantities. Following necrosis, regeneration of cells occurs in some of the organs (liver, pancreas, kidney).

A second result is scleroses which are formed in the liver, pancreas and heart, owing to the disappearance of the parenchymatous cells in places and the subsequent shrinkage and coalescence of the stroma. These secondary lesions are recognized as cirrhosis of the liver, which may cause ascites and jaundice, and selerosis of the pancreas, which if severe enough always produces diabetes mellitus and sclerosis of the heart.

The third clinical sign, pigmentation of the skin, may be due to the primary lesion, *i. e.*, the deposition of hemofuscin in certain cells in the corium followed by transformation to hemosiderin or to increase of melanin in the epidermis owing to injury to the adrenal gland (deposition of pigment in the zona glomerulosa) or to the combination.

Pathogenesis. The etiology of hemochromatosis has always been a fertile field for discussion because nothing definite is known about it. The first theory advanced in explanation of its origin was that the condition was secondary to the diabetes, the second that it followed the cirrhosis of the liver. These two theories have been very generally discredited because plenty of instances of both types of lesions occur without any sign of pigmentation. Recently, however, Rous and Oliver, in their paper on "Experimental Hemochromatosis," have again advocated a primary cirrhosis of the liver as the starting-point in causing a failure of the organ to deal adequately with the iron-containing products of normal blood destruction, so that they accumulate in the body and cause automatically widespread pigmentation.

The third theory, first advanced by Marie, in 1895, is that the

lesions are due to some as yet unknown toxin which is responsible for the hemosiderosis. The deposit of pigment then secondarily produces the sclerosis of the liver and pancreas.

The fourth theory would explain the source of the pigment as due to a perverted iron metabolism, as a result of which the cells of the various organs and tissues retain most of the iron which reaches them through the circulation. The deposition of the pigment is regarded here also as the primary lesion which in time leads to sclerosis of the liver and pancreas.

The importance of the third theory will appear in connection with the results obtained by experimental work to be presented later. The great objection to it has been the apparently necessary great destruction of red blood corpuscles to account for the large amount of iron deposited in the tissues; of this no evidence has ever been obtainable either clinically or postmortem.

Technical Methods Employed. The histological study of the lesions described here has been based largely on paraffin sections of Zenker-fixed tissues stained by the eosin-methylene blue method. The demonstration of the two kinds of pigment granules, hemofuscin and hemosiderin, however, required special methods. Inasmuch as the chemical properties and staining reactions of these pigments are of much importance in the study of hemochromatosis a brief statement of them seems advisable.

Hemofuscin occurs in the form of pale yellow granules, of various sizes up to a limited maximum, which are soluble in alkalies and in peroxide of hydrogen but not in dilute acids. They stain more or less deeply with basic anilin dyes, especially methylene blue, fuchsin and safranin, but do not stain with alum hematoxylin. The method found most useful for the demonstration of them is as follows:

Directions. Fixation in Zenker's fluid, alcohol or formaldehyde.

1. Stain paraffin sections in alum hematoxylin until the nuclei stand out sharply defined.
2. Wash in water.
3. Stain for ten to thirty minutes in the following solution:

Fuchsin	0.5 gm.
Alcohol, 95 per cent	50.0 cc.
Water	50.0 cc.

4. Wash off in water.

5. Differentiate and dehydrate in 95 per cent followed by absolute alcohol.

6. Xylol, xylol colophonium.

Nuclei blue, hemofuscin granules bright red, hemosiderin unstained.

The method can be used equally well on celloidin sections if oleum origani cretici is employed in place of xylol.

The stain differentiates sharply the granules of hemofuscin from those of hemosiderin, but no other claim is made for it except that it does not stain melanin.

Hemosiderin occurs in the form of yellowish or orange-colored granules which often have a brownish tint, especially when occurring in masses. They are soluble in dilute acids but are not affected by alkalies or by peroxide of hydrogen. They are not stained by anilin dyes or by alum hematoxylin. On the other hand they give the characteristic color reactions of iron with ammonium sulphide and with ferrocyanide of potassium and hydrochloric acid. Apparently the hemosiderin in hemochromatosis always occurs as a ferric salt.

The following methods will be found to give excellent results:

A. *Reaction with Ammonium Sulphide.* Fixation in Zenker's fluid, alcohol or formaldehyde.

1. Stain paraffin sections in alum hematoxylin until the nuclei are of a deep blue color.

2. Wash in water.

3. Place in a mixture of one part of strong yellow ammonium sulphide to three parts of 95 per cent alcohol for one or two hours or longer. Use a glass staining dish with tightly fitting cover to prevent evaporation of the ammonia.

4. Wash quickly but thoroughly in several changes of water.

5. Dehydrate in 95 per cent and absolute alcohol.

6. Xylol, xylol colophonium.

Nuclei blue, hemosiderin granules of an intense black. The stain keeps well. The results after the Zenker fixation are perfect. If the sections are kept in water or alcohol too long the black color fades as the sulphide changes to the hydroxide.

The drawback to the ammonium sulphide reaction for general purposes is, of course, the fact that other metals besides iron form black sulphides, but this point is negligible in hemochromatosis, and if necessary can always be controlled by the Berlin blue reaction. The resulting stain is exceedingly sharp even with tissues preserved for years in formaldehyde, and is especially recommended for photographic purposes, as it yields results which are like etchings.

B. *Berlin Blue.* Due to the action of ferrocyanide of potassium and hydrochloric acid on a ferric salt.

This is the classic diagnostic color reaction for iron due to the formation of Berlin blue. It has at least two drawbacks: Hemosiderin is soluble in the hydrochloric acid and the Berlin blue in the ferrocyanide of potassium. As a result the stain readily diffuses, the fine granules all disappear and the larger ones have blue halos around them. A fine precipitate may be formed and deposited on the surfaces of the sections.

There are various methods of performing the test. The following is recommended for ordinary purposes:

Fixation in alcohol or formaldehyde. After Zenker's fluid the hemosiderin dissolves much too easily in the hydrochloric acid, so that only a diffuse stain results.

Directions. 1. Stain paraffin (or celloidin) sections for ten to twenty-five minutes in the following mixture:

Ferrocyanide of potassium, 2 per cent aqueous solution, freshly prepared (not over one week old)	1 part
Hydrochloric acid, 1 per cent aqueous solution	3 parts

Better results will be obtained if, following a suggestion by Maude E. Abbott, the reaction is hastened by high temperature. Heat the mixture in a test-tube until beads of gas form on the inner surface of the glass (about 80° to 85° C.) and pour over the sections. The reaction takes place within thirty to forty-five seconds.

2. Wash thoroughly in several changes of water.

3. Counterstain lightly in alum carmin or in a one-tenth per cent aqueous solution of safranin for two to five minutes.

4. Wash off in water.

5. Differentiate and dehydrate in 95 per cent followed by absolute alcohol.

6. Xylol, xylol colophonium.

Nuclei red, hemosiderin blue, hemofuscin pale red.

In the method recommended here the reaction takes place so completely, owing to the excess of acid, and so quickly, owing to the heat, that there is little time for diffusion. The stain is intense and sharp except for the finest granules.

C. *Turnbull's Blue.* The second diagnostic stain for iron is due to the action of ferricyanide of potassium and hydrochloric acid on a ferrous salt.

When hemosiderin is treated with ammonium sulphide the finest granules and the surface of the coarse ones are changed to ferrous sulphide. In consequence of this transformation sections first stained with ammonium sulphide may be treated with ferrocyanide of potassium and hydrochloric acid (MacCallum, Hall, Nishimura) and give Berlin blue, or with ferricyanide of potassium and hydrochloric acid (Tirmann) and give Turnbull's blue, but neither method can stain all the iron present. Moreover, for the reasons already stated, both these methods are faulty and imperfect on account of the diffusion of the blue color, owing to the action of the hydrochloric acid. If, however, the acid is omitted in the second reaction the ferricyanide of potassium alone will give a perfect deep blue stain after ammonium sulphide, without diffusion, provided its time of action is limited, which rivals the ammonium sulphide stain in sharpness and surpasses it in color and diagnostic value. The intensity of the blue stain depends on the length of action of the ammonium sulphide. If it is short only the surface of the coarse granules is transformed to ferrous sulphide. When the color is turned blue the yellow within shows through the blue surface stain, making the larger masses appear dark greenish. Therefore it is important to have the reaction with ammonium sulphide as complete as possible. All the hemosiderin, so far as practicable, should be turned to a ferrous salt. The reaction is quicker and more effective and takes place even after Zenker fixation if 5 per cent of acetic acid, which seems to have no dissolving effect on either hemosiderin or Turnbull's blue, is added to the ferricyanide of potassium.

Directions. Alcohol, formaldehyde or Zenker fixation.

1. Stain paraffin sections in—

Strong yellow ammonium sulphide	1 part
Alcohol, 95 per cent	3 parts

for one or two hours or longer (twelve to twenty-four hours for Zenker fixed tissue).

2. Wash off in water.

3. Place in a freshly prepared mixture of—

Ferricyanide of potassium (2 per cent aqueous solution)	19 cc.
Glacial acetic acid	1 cc.

for ten to twenty minutes or longer (twelve to twenty-four hours for Zenker fixed tissue).

4. Wash thoroughly in several changes of water.

COUNTERSTAINS. (*a*) *For Ordinary Tissues Where Only the Iron Reaction is Desired.* Lithium carmin, which is ordinarily recommended, is inadvisable because of the injurious effect of the alkali in it and of the acid following it. Use alum carmin or a dilute solution of safranin or fuchsin.

(*b*) *For Hemochromatosis Material in Which it is Desirable to Stain both Kinds of Pigment.* Use the stain for hemofuscin, steps 3 to 6. The alum hematoxylin is omitted because it dulls the picture. Hemosiderin blue, hemofuscin brilliant red, nuclei red, especially after Zenker fixation.

PREVIOUS ATTEMPTS TO PRODUCE HEMOCHROMATOSIS. Various attempts to produce hemochromatosis experimentally all seem to have been based on the idea that the hemosiderin is derived from hemoglobin set free by an abnormal destruction of red blood corpuscles. Two lines of experiments have been tried: (*a*) The injection of blood into the peritoneal cavity to represent hemorrhage, and (*b*) the use of poisons, such as toluylenediamin, which destroy the red blood corpuscles in the circulation.

The most promising work along this line has been reported recently by Rous and Oliver. They injected large amounts of rabbits' blood into the circulation of rabbits almost daily for many months, and as a result obtained deposits of hemosiderin in the liver,

pancreas, heart, spleen and other organs and tissues. They noted slight cirrhotic changes in the livers of two rabbits which had survived a long time, but interpreted them as of an intercurrent nature and not due to the siderosis. Their conclusion is that while their results do not indicate the ultimate cause of the disease they are practically identical with the siderosis characterizing hemochromatosis and throw light on its various features and on its course.

Experimental Pigment Cirrhosis Due to Chronic Poisoning with Copper. The lesions to be described under this heading are an outcome of a number of years of experimental work with various chemical reagents which might possibly be the cause of the two chronic and in some ways closely related toxic conditions known as alcoholic cirrhosis and hemochromatosis. It was and is believed that there is a perfectly definite etiologic agent for each of them.

If half a gram of acetate of copper is given by stomach tube to a rabbit weighing two or three kilos it will cause death in less than twenty-four hours. If the dose in the beginning is made 0.1 gram it can be gradually increased in the course of a week to 0.2 gram and carried along at this amount daily without causing the animal any discomfort. A simpler way is to mix a solution of the chemical with the food. A rabbit will readily take about 0.1 gram a day in this way, and this amount was found to be sufficient to cause cirrhosis of the liver and death in from six to twelve months. For more chronic lesions a smaller dose and longer time are required.

The following general description is based on a series of twenty-two rabbits which died or were killed at various periods of time from one week up to eleven and a half months. The primary lesion was the same in all of them, depending only on the size of the dose and the length of time the copper was administered.

In a very short while, one to three weeks, small yellow pigment granules begin to appear in the endothelial cells lining the sinusoids and in the liver cells. In one to two months they are numerous, in three months abundant. All the liver cells contain them, but especially those at the peripheries of the lobules. In four or five months the condition is still more marked. Many of the liver cells have been killed as the pigment has increased in amount and other liver cells have regenerated. When a cell undergoes necrosis it is

invaded by polymorphonuclear leukocytes which remove the cytoplasm. The pigment granules are taken up by endothelial leukocytes which stuff themselves full and tend to migrate to the periportal connective tissue and collect there in large numbers in the lymph vessels and between the fibroblasts around the blood-vessels and bile ducts. The regeneration of liver cells in time becomes very active. As many as five mitotic figures (monasters and diasters) have been found in one oil-immersion field. Occasionally triasters occur. The newly formed cells usually contain little or no pigment while the old ones are full of it.

The first increase of connective tissue appears in the neighborhood of the portal vessels where the liver cells are most pigmented and in consequence necrose first. As the cells disappear the elements of the stroma left behind coalesce and form foci of sclerosis. Where liver cells regenerate new stroma is formed for them.

Occasionally pigment granules were found in fibroblasts, but there was never any evidence that they caused necrosis of the cells.

The earliest case of definite cirrhosis occurred at the end of five and one-half months. The liver was somewhat enlarged and noticeably dense to the touch. The surface was slightly and finely granular. Pieces of the tissue fixed in alcohol stained it yellow, indicating bile stasis, and this was confirmed by microscopic examination.

The second case of cirrhosis came at the end of six and one-half months and was accompanied by intense jaundice of all the organs and tissues of the body.

A third animal died at the end of eleven and a half months with cirrhosis and also with intense jaundice.

Microscopic examination of all three cases of cirrhosis showed well-marked increase of connective tissue not only around the portal vessels but also running in various directions within the lobules.

All the pigment granules in this series of twenty-two rabbits have the chemical properties and the staining reactions of hemofuscin. None of them has changed to hemosiderin. Their presence gives to the liver on fresh examination a chocolate-brown color, very pronounced after poisoning which has lasted for several months,

except so far as it is modified to a brownish or greenish yellow in the cirrhotic cases by the presence of jaundice.

Outside of the liver pigment granules were found in endothelial leukocytes in the mesenteric lymph nodes and to a slight extent in the spleen and bone-marrow. A few were present in the endothelial cells lining the capillaries of the heart but none in the cardiac muscle cells or in the epithelium of the pancreas.

The examination of the blood was at all times negative. There was no evidence of anemia such as occurs so characteristically in lead-poisoning, owing to destruction of the red blood corpuscles.

Feeding acetate of copper to other animals than rabbits gave the following results:

The deposition of pigment in the livers of guinea-pigs takes place very much slower than in the rabbit. On this account work carried on about eight years ago with this same chemical, but fed only to guinea-pigs, yielded no results. It requires over a year to produce a moderate degree of sclerosis.

White rats show pigment in the liver about as quickly as rabbits do, but have not been carried far enough along to develop cirrhosis.

A ring-tail monkey which died at the end of five and a half months' feeding with a small amount of acetate of copper on its food showed a fair degree of pigmentation of the liver cells. About half the cells stained like hemofuscin. The rest gave a distinct iron reaction. Apparently the liver cells of the monkey possess to a much greater extent than do those of the rabbit the power to break down the copper hemoglobin compound and form hemosiderin.

A second monkey, rhesus macacus, fed on smaller doses is still alive at the end of fifteen months.

Of the other metals which form hemol compounds with hemoglobin, zinc is the only one which causes a lesion closely resembling that produced by copper.

Two other lines of experimentation with copper were tried. Chloride of copper in small doses was injected directly into the circulation of a rabbit for a period of thirty-three days. The same result was obtained as when a copper salt was introduced into the gastrointestinal tract, namely, pigmentation of the cells of the liver but not of those of other organs. This experiment would seem to

show definitely that liver cells take up the hemol compound more quickly than do other parenchymatous cells, not because it reaches them first but because of some special attraction.

In like manner a compound of copper with hemoglobin forming cuprohemol was injected into the circulation of two rabbits. The only parenchymatous cells in which the pigment appeared were those of the liver. This line of experimental work was not continued because it did not seem promising and the animals were easily killed if a very small dosage was exceeded.

While acute poisoning with copper is well recognized the general consensus of opinion is that chronic poisoning does not occur except occasionally among workers in this metal, and even this, according to some observers, may be due to the zinc present in brass. The symptoms caused point chiefly to the respiratory tract. Halsey in his translation of Meyer and Gottlieb's *Pharmacology* goes so far as to state in an insert, "The prohibition of copper as a coloring agent for foods is not justifiable from a hygienic standpoint."

On the other hand some investigators have looked on copper with suspicion, and considerable experimental work on animals and even on man has been done with practically negative results. The criticisms of the work recorded are two: The administration of the copper was not continued a long enough time to obtain results, and the work apparently was not carefully controlled by microscopic examination.

These criticisms apply only in part, however, to the experimental work of Baum and Seeliger. They poisoned with various salts of copper a number of animals of different kinds (goat, sheep, dog, cat) for varying periods up to almost a year and observed "hemoglobin derivatives (hematoidin and hemosiderin)" in liver cells. They also noted the frequent presence of masses of hemosiderin in the liver but failed to recognize the early stages of sclerosis. They ascribed the pigments to the poisonous effect of copper but made no attempt to explain how they got there and very evidently did not realize their significance, that is, their possible relation to hemochromatosis and to the cirrhosis occurring in that disease. The animals they worked with, like the monkey used by us, apparently are able to break down the cuprohemol compound, so that

hemosiderin is formed. The results obtained by them, therefore, still more strongly favor the view set forth by us that chronic poisoning by copper may be the cause of hemochromatosis.

If hemochromatosis is due to chronic poisoning with copper, as the experimental work detailed here strongly suggests, the important question is, How does it obtain entrance to the body? There are several sources.

The salts of copper have long been used in connection with certain foods on account of their coloring properties. "To copper" is to stain bright green with these substances. The foods most often colored are pickles, canned peas and beans. The old-fashioned way was to use a copper kettle and a little vinegar, but the salts have often been added directly. In France, where hemochromatosis is relatively common, they are often added to absinthe to improve its color.

The salts of copper are also used to some extent on account of their antiseptic properties. Thus they are added to bread, beer and wine to prevent souring and to fresh water to stop or limit the growth of algæ during the summer months.

Still another, and possibly the most important source of poisoning, may be from distilled liquors. Copper or brass tubing is often used in the coils and pipes connected with the still instead of block tin. The chief acid in the mash is acetic. This passes over along with the other volatile substances and readily attacks copper, as is easily demonstrated experimentally.

Alcohol is claimed by various investigators to bear an intimate relation to hemochromatosis.

Discussion. *Hemochromatosis and Alcoholic Cirrhosis.* In studying the gross and microscopic appearances of cirrhosis of the liver as found in hemochromatosis it is important to bear in mind that, as already stated, the condition is often complicated by the lesion of alcoholic cirrhosis, and *vice versa*. Histologically the two types of lesions are entirely distinct: one is characterized by the presence of pigment in the liver cells, and the other, alcoholic cirrhosis, by a fine to coarse hayaline meshwork in affected cells. Both types of lesions may occur not only in the same liver but sometimes in the same cells. For purposes of discussion or descrip-

tion, livers with but a single type of lesion should be selected. The three cases described by Sprunt, from whom material was available for study, were all pure types of hemochromatosis.

The frequent association of the two types of lesions in the same liver suggests an analogous cause for each.

Hypertrophic and Atrophic Cirrhosis. The size of the liver in hemochromatosis depends, as in alcoholic cirrhosis, on the intensity and duration of the process. If the lesion is active and early it may cause increase in the weight of the liver up to 2500 grams or over (3200 grams in one of Sprunt's cases), owing to deposit of pigment, inflammatory infiltration with leukocytes to remove necrotic liver cells and the pigment, regeneration of liver cells and formation of new stroma. The surface may be smooth, slightly irregular or granular, the edges rounded and the density more or less increased. The color is light to dark reddish brown, or black if affected by postmortem changes.

If the process has been mild or intermittent and has lasted a great many years, regeneration in time fails to equal the rate of destruction and the liver diminishes in size to below normal. It may weigh as low as 1020 grams. The surface is very irregular and nodular and presents the typical hobnail appearance so characteristic of alcoholic cirrhosis in its late stages. The granules and nodules represent foci of regeneration; the depressions, contracted bands of old connective-tissue stroma. The density is greatly increased and the color is light to deep bronze, depending on the degree of pigmentation, which may be only moderate because endothelial leukocytes may have digested and removed all the hemosiderin set free by the necrosis of liver cells.

Extracellular Hemosiderin. The statement is often made, in descriptions of the lesion of hemochromatosis, that more or less of the pigment is extracellular. Unna used to make the same claim about leprosy bacilli. The statement in both instances is probably incorrect.

Immediately following necrosis of parenchymatous cells filled with pigment the cytoplasm is dissolved and removed first. Then the hemosiderin, which exerts a strong chemotactic attraction on endothelial leukocytes, is immediately taken up by them in large

quantities. Rarely two or more of the leukocytes fuse together to form foreign body giant cells. The pigment persists indefinitely in these cells until it is digested and returned in some form or other to the circulation to be disposed of.

Rounded and oval masses of pigment not in epithelial cells mean endothelial leukocytes filled with it, although the cell outlines and nuclei are often more or less completely obscured. Rows of granules that seem to be free are always contained in flattened endothelial cells or in the cytoplasmic processes of fibroblasts.

Source of Pigment. One of the puzzles connected with hemochromatosis has always been the origin of the pigment, which is evidently derived from hemoglobin. It has been very generally ascribed to an increased destruction of red blood corpuscles, but no evidence of such destruction has ever been found clinically or postmortem, and the spleen and bone-marrow do not present the changes which are characteristic of such destruction.

The same peculiar condition is true of animals poisoned with copper. Examination of the blood shows no destruction of red blood corpuscles. There is no enlargement of the spleen until after cirrhosis has occurred and no abnormal activity of the bone-marrow.

What is the explanation? It is probably very simple, namely, that there is no abnormal destruction of red blood corpuscles.

According to the physiologists one-twentieth to one-twelfth of the red blood corpuscles in the body disintegrate daily under normal conditions. The hemoglobin set free in this way circulates in the blood and is eventually eliminated. Copper has a great affinity for free hemoglobin (Kobert) and unites with it to form a compound known as cuprohemol. Apparently in the circulating blood it does not unite with the hemoglobin within the red cells and therefore does not cause destruction of them. Cuprohemol, or a derivative of it, is deposited as a yellowish pigment in the cells of the liver and leads eventually, as already described, to cirrhosis.

Diminished Elimination of Iron. It has been demonstrated by Howard and Stevens, and confirmed by McClure, that the elimination of iron in hemochromatosis is lowered. In other words, iron is retained in the body. This retention is readily explained on the supposition that the hemoglobin set free by the normal disintegra-

tion of red blood corpuscles unites in the circulation with copper absorbed from the intestinal tract and is deposited as hemofuscin in the cells of the liver and in other organs and tissues of the body. Most of the cells readily break down this primary pigment into hemosiderin.

Proof that Hemochromatosis is Due to Chronic Poisoning with Copper. The experimental work reported here suggests strongly that hemochromatosis may be due to chronic poisoning with copper. The proof that copper is the cause is quite another thing, and proof is not easy to obtain.

The amount of hemofuscin present in the body at any one time in a case of hemochromatosis is very small, probably much less than one-hundredth part of the iron; therefore the possible amount of copper presumably is very slight also.

An excess of copper in the body might be demonstrated in at least three different ways:

(*a*) *Copper in Hemofuscin.* This would require showing the presence of copper in the hemofuscin granules in the cells by means of microchemical tests. This is no more possible than demonstrating the iron also present in the cuprohemol compound. Both the copper, if present, and the iron are so intimately bound up in the organic compound that no diagnostic reaction can be obtained.

(*b*) *Copper in the Liver.* By using large amounts of liver tissue and digesting it with strong acids it would be possible to demonstrate copper in the same way that iron can be shown to exist in hemoglobin. The test, however, would require tissue in large amounts, and this has not been available.

(*c*) *Elimination of Copper.* It seems probable that copper if present in hemofuscin would be eliminated when that substance is broken down to form hemosiderin. This offers a third method, and perhaps the best, of seeking evidence whether or not hemochromatosis is due to chronic poisoning with copper.

We have tried the first and second methods without success. A favorable, that is early, active case of hemochromatosis in the hands of a good analytical chemist might throw light on the subject.

Increase of Normal Body Pigments. It is commonly stated that in hemochromatosis there is a general increase of pigments through-

out the body in cells which normally contain it. Opie ascribes the pigmentation of the skin entirely to an increase of melanin in the epidermis. If this is true it is possible that it is caused by the injury to the outer layer of cortical cells, the zona glomerulosa, of the adrenal glands, and that the skin lesion really is one variety of Addison's disease. In other words, pigmentation of the skin may follow injury to the adrenal glands, owing to the deposition of pigment, in the same way that ascites follows cirrhosis of the liver and diabetes mellitus the sclerosis of the pancreas.

In marked amyloid formation of the adrenal glands only the zona glomerulosa is spared. Practically all the rest of the parenchymatous tissue is destroyed by the pressure due to the amyloid, and yet no pigmentation of the skin occurs.

Clinically it is well recognized that the pigmentation of the skin due to Addison's disease and to hemochromatosis cannot be positively differentiated from each other except by the aid of the existence of other signs (ascites and diabetes mellitus) or most conclusively by the demonstration of hemosiderin in the skin.

The hemofuscin deposited in cells of the corium and the hemosiderin derived from it may or may not play some part in the appearance of pigmentation. It is rather deeply placed to produce much effect, and yet the silver in argyria occupies about the same location. The question of pigmentation of the skin cannot be decided solely by postmortem examination. It is most important to know if the patient was originally of the blonde or brunette type.

Poisoning with Iron. Ferric chloride in 1-gram doses was administered to a rabbit daily by stomach tube for five and a half months. On postmortem examination the liver was of a bright reddish-brown, rust-like color. Microscopically the tissue showed the deposition of much iron in the form of yellow granules. It was in endothelial and liver cells and occurred most abundantly at the peripheries of the lobules. Occasional liver cells had undergone necrosis and the pigment had been taken up by endothelial leukocytes. It seems probable that if the administration of iron had been continued long enough cirrhosis would have resulted.

In the experimental work of Rous and Oliver, where rabbits' blood was injected into the circulation of rabbits for many months,

two out of the three animals which survived longest showed moderate cirrhosis of the liver. The authors interpreted the condition as of an intercurrent nature. It is probable that it was produced by the deposition of hemosiderin and that the third rabbit in time would have shown the same lesion. Animals are not equally susceptible to injurious agents. The diagnosis of the cirrhosis should have been based on necrosis and regeneration of liver cells. If those two processes were in evidence the cirrhosis in two of the rabbits was due to the hemosiderin and in time would have occurred in the third.

The theory of Rous and Oliver that cirrhosis of the liver is the primary condition which leads to retention of iron-containing products of normal blood destruction, so that they accumulate as pigment in the various organs and tissues of the body, is untenable in view of the many cases of all degrees of cirrhosis of the liver of alcoholic and acute yellow atrophy types which occur without a trace of hemosiderin being found in the liver or elsewhere. They also fail to consider the acute early cases of hemosiderosis of the liver in which little or no cirrhosis has yet occurred.

Susceptibility. The experimental work with copper has shown quite definitely that all rabbits are not equally susceptible to its poisonous effects. Even animals of the same litter failed to develop lesions with equal rapidity. On the same dose one rabbit died from cirrhosis in five and a half months, a second in six and a half months and a third in eleven and a half months.

When we compare the susceptibility of different kinds of animals the difference is still more marked. Rabbits, white rats and monkeys are all fairly susceptible to poisoning by copper; guinea-pigs, on the other hand, are very slightly so.

Human beings may differ much in their reaction to copper just as, it is well recognized, they do to arsenic, lead and some other poisons.

Cause of Necrosis of Parenchymatous Cells. In hemochromatosis the parenchymatous cells which undergo necrosis are those which are overloaded with hemosiderin. Hemofuscin never seems to be present in sufficient quantity to cause injury. In the experimental animals, on the other hand, the cells which die are those filled with

cuprohemol or a derivative of it. Only the more highly differentiated cells, such as liver, pancreatic and cardiac muscle cells, succumb. Fibroblasts and endothelial cells and leukocytes persist indefinitely filled with either kind of pigment.

Necrosis does not seem to be due to a chemical action on the part of either kind of pigment but rather owing to mechanical action, interfering, perhaps, with the normal functions of the cells.

Primary Carcinoma of the Liver. Primary cancer of the liver is comparatively rare. On this account its relatively frequent occurrence in combination with cirrhosis is of interest. In our two hundred and twenty-four cases of cirrhosis primary cancer of the liver was present four times, once associated with the hemochromatosis type of lesion, three times with the combined lesions of alcoholic cirrhosis and hemochromatosis. In addition specimens from two other cases of carcinoma of the liver combined with hemochromatosis have been sent to the laboratory during this past year. The tumors all seemed to be of the primary liver-cell type, although the cells in some of them were not fully differentiated. In one instance metastases to regional lymph nodes had taken place. One of the four cases of hemochromatosis recently reported by Blanton and Healy had a primary cancer of the liver.

The significance of these cases is that they would seem to show some definite relation between long-continued active regeneration of liver cells and primary carcinoma of the liver.

Summary. Hemochromatosis is a chronic disease due to a definite toxic agent which unites with the hemoglobin set free by the normal disintegration of red-blood corpuscles. From the compound thus formed hemofuscin is deposited in various cells throughout the body. It appears first and in largest quantities in the liver; later in the pancreas, heart, kidneys, adrenal glands and other organs and tissues. Most cells (endothelial, hepatic, pancreatic, etc.) readily transform the hemofuscin so that hemosiderin results, as can be readily demonstrated by the iron reactions. Other cells (epithelium in ducts, smooth muscle cells, fibroblasts in certain locations) have little or no effect on the hemofuscin, so that it persists in that form more or less indefinitely.

The effect of the excessive deposit of pigment in parenchymatous

cells (liver, pancreas, heart) is to cause necrosis of some of them followed by active regeneration by others in certain organs (liver, pancreas). The pigment set free by necrosis is taken up in large quantities by endothelial leukocytes which collect, sometimes in great numbers, everywhere in the stroma.

The connective tissue in some of these organs, liver for instance, is gradually increased owing to new formation of fibroblasts, as in tumors, to form stroma for the islands of regenerated cells. The sclerosis is due to contraction and coalescence of old stroma as the pigmented cells die off.

The final result of these processes is cirrhosis of the liver, sclerosis of the pancreas with consequent diabetes mellitus and sclerosis of the heart. The pigmentation of the skin may be due, in part at least, as in Addison's disease, to injury to the adrenal glands (zona glomerulosa).

The spleen and bone-marrow show no marked changes of any sort and clinically there is no evidence of anemia.

Chronic poisoning with salts of copper produces in rabbits within six to twelve months well-marked cirrhosis of the liver with intense jaundice followed quickly by death. The liver appears of a dark chocolate color owing to the deposit of great numbers of yellow pigment granules in the liver and endothelial cells most marked at the peripheries of the lobules. Excessive deposit of pigment results in necrosis of liver cells followed by active regeneration.

In the rabbit the pigment, which has the same staining peculiarities as hemofuscin, does not undergo transformation into hemosiderin. In a monkey, however, which died after copper-poisoning of five months' duration over half of the pigment granules in the liver cells gave a definite iron reaction.

The only deposit of pigment outside of the liver, except for occasional pigmented endothelial leukocytes in the spleen and bone-marrow, occurred in the kidney and in endothelial cells lining some of the capillaries of the heart.

The spleen and bone-marrow showed practically no changes until after cirrhosis had occurred, and examination of the blood for evidence of the destruction of red blood corpuscles was always negative.

Conclusions. It is impossible to produce in months lesions for which Nature requires years.

Chronic poisoning with salts of copper produces in the livers of rabbits in six months to a year a series of changes comparable in many ways with those found in the liver in a chronic disease in man known as hemochromatosis. Poisoning with smaller doses continued over a much longer time would probably cause lesions in other organs.

Proof that hemochromatosis is due to poisoning with copper would require the demonstration of copper either in hemofuscin or in the liver in excess of the minute amount said to be normally present or in excretions from the body.

If hemochromatosis is due to poisoning with copper there are several sources from which it may be derived:

1. From its use as a coloring reagent to "copper" vegetables (pickles, canned peas and beans) and to improve the color of absinthe.

2. From its use as an antiseptic to prevent fermentation (beer, wine) or to inhibit the growth of algæ (drinking water).

3. From distilled liquors (action of acids, chiefly acetic, on copper and brass tubing used in connection with stills).

The present extensive use of crude distilling apparatus in consequence of prohibition is likely to lead to an increase in the number of cases of hemochromatosis if the disease is due to chronic poisoning with copper.

BIBLIOGRAPHY

Abbott, Maude E.: Pigmentation Cirrhosis of the Liver in a Case of Hemochromatosis, Jour. Path. and Bacteriol., 1901, vii, 55.

Opie, E. L.: A Case of Hemochromatosis; the Relation of Hemochromatosis to Bronzed Diabetes, Jour. Exper. Med., 1899, iv, 279–306.

Sprunt, T. P.: Hemochromatosis, Arch. Int. Med., 1911, viii, 75–129.

Rous, Peyton, and Oliver, Jean: Experimental Hemochromatosis, Jour. Exper. Med., 1918, xxviii, 629–644.

Baum and Seeliger: Die chronische Kupfervergiftung, Arch. f. Thierheilk., 1898, xxiv, 80–127.

Howard, C. P.: Hemochromatosis, Oxford Med., 1921, iv, 215–222.

An extensive bibliography will be found in each of the above papers.

Blanton, W. B., and Healy, W.: Hemochromatosis, Report of Four Cases, Arch. Int. Med., 1921, xxvii, 406–420.

Marie, P.: Sur un cas de diabète bronzé suivi d'autopsie, Sémaine méd., 1895, xv, 229.

Howard, C. P., and Stevens, F. A.: The Iron Metabolism of Hemochromatosis, Arch. Int. Med., 1917, xx, 896.

McClure, C. W.: Metabolism in a Case of Hemochromatosis, Arch. Int. Med., 1918, xxii, 610.

DISCUSSION

Dr. N. E. Brill, New York: Did the reader of the paper consider the question as to whether there might not be some chemical or biologic differences in the pigments, such as those which result from hemolytic conditions, such as one, for example, which is associated with pernicious anemia, in which the hemoglobin of the cells is subject to disintegration, and the pigments on the other hand resulting from a non-hemolytic disease such as hemochromatosis? We know that in the latter disease there is no destruction of the red cells such as occurs in the former, that it is unaccompanied by any indications of anemia or by any elements of splenic or bone-marrow activation. The fact that there is pigmentation occurring in both these entirely dissimilar diseases might suggest that there might be some differences in the characters of the pigments of each.

Dr. L. G. Rowntree, Rochester, Minn.: Did Dr. Mallory see any evidence of toxicity from the copper salts? Were there any blood changes?

Dr. T. Sollmann, Cleveland, Ohio: I should like to understand the effect of copper salts on the blood pigment?.

Dr. F. B. Mallory (closing): Copper salts do not affect the red-blood corpuscles in the circulation at all. In this respect they are quite different from those of lead which cause destruction of them. According to physiologists a large number of the red-cells are destroyed daily, so there must be free hemoglobin in the circulation. The copper salts unite with this free hemoglobin to form cuprohemol. This pigment, or a derivative of it, is deposited in the liver cells and acts as an injurious agent causing necrosis and eventually cirrhosis. There is no enlargement of the spleen and no effect is produced on the bone-marrow.

PARASITISM AS A FACTOR IN DISEASE

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The study of etiology or causation is a study of the entire field of medicine from a certain point of view. Every phenomenon assumes an etiologic aspect whenever we study it, not as an effect to be simply contemplated and described but as a cause or necessary condition of something that is going to happen. Provided with the information that for certain events to take place certain necessary conditions must precede, we can take steps by controlling the necessary conditions to allow the event to occur or not. Modern medicine has made the concept of causation its own. On it is founded all rational progress in prophylaxis and therapy. First to comprehend the cause, then to intercept and suppress it, and thereby to prevent the next step, is the kernel of medical science and practice. We project ourselves into the immediate future. The present is only the boundary between what has occurred and what is to happen. To control events we must know how to distinguish those conditions which are necessary from those which are merely associated and coincident.

The history of medical science, notably during the past half-century, has clearly shown that observation of disease as it occurs in every-day life must be associated with the experiment. By observation I mean a survey or study of the phenomenon as a whole; by experiment, the observation of isolated parts of the entire phenomenon, the other parts being meanwhile eliminated or controlled by special devices. Observation and experiment, alternating, coöperating and reacting on each other, are the only sure guides to a rational interpretation of disease. Nature is continually experiment-

ing and observation is simply taking notes in this great life-experiment. Without it the laboratory experiment would lack reality, for it is simply a page torn out of the book of Nature, and to become useful it must be fitted in again. Experiments are imitations of Nature with the unknown factors controlled or eliminated. To get at the facts of disease it has been found necessary to bring experiments as close as possible to the natural phenomena without losing control of the details.

The necessary association of observation and experiment in interpreting the conditions determining disease may be illustrated by the familiar one of the noise and the flash of gunfire. At a distance we see the flash and then hear the explosion. We might infer that the flash was a cause of the explosion, since it always precedes it. As we approach the scene of operations the noise follows the flash more and more quickly and close by the two reach our senses almost simultaneously. We then are in doubt whether the flash is a cause or merely an accompanying phenomenon. Our static observation at a distance fails to inform us correctly. The experiment of approaching the firing compels us to revise our original notion of causation and to make a further study of the entire phenomenon. The gist of the etiologic problem is thus to determine what are necessary conditions and what merely secondary phenomena. The experimental method has been of immense service in laying bare the dynamic or causal relation—in other words, the true sequence of events. On the other hand, experiment too far removed from the actualities has frequently led astray when its results were too literally accepted and not controlled by observation of the entire phenomenon.

In the environment of man, and within the human mechanism itself, there are many conditions operative toward disease. This entire group or sequence of conditions, rather than any single factor in the group or sequence, may be regarded as the cause. If any one of these conditions is neutralized or controlled the disease may not occur, or if in progress it may take another course. Naturally these conditions have different values. They may be judged from their accessibility to control, *i. e.*, from a practical standpoint or from the point of view of the physicist weighing them according to

their energy values. We have hardly reached the stage, however, when the conditions favoring disease can be accurately measured. We must still deal with them as entities. Their qualities must engross our attention and their quantitative relations remain for a future and more exact medical science to weigh and measure.

The forces and conditions controlling disease are a mixture of heredity, environment and parasitism. How can these factors be taken from their natural relations and studied individually without upsetting the delicate balances of causation? Where can we begin to test experimentally the observations we make about natural occurrences? Obviously, some very careful surgical operation is necessary in carving out our field of work. In so doing we must realize that we become piece-workers, tinkering with only a part of Nature's mechanism. Our finished product must be skilfully fitted into the larger mechanism. In attempting to limit our discussion to parasitism as an etiologic factor in disease I realize the difficulties mentioned. We have not only the different categories of environment, heredity and parasitism acting on one another, but within each category we have the animal body reacting with the factors like a chemical process swinging back and forth toward a state of equilibrium. Finally, we have in parasitism two living variable organisms capable of adjusting themselves toward each other in a remarkable degree.

When about forty years ago methods were devised by Robert Koch to make a beginning in the accurate study of bacteria as living agents of disease, the contemporary scientific world realized that here were, to all appearances, agencies that could be separated from their environment, their life-history and activities subjected to rigid investigation and their relation to disease opened to demonstration. It is not surprising, therefore, that the bacteriologists of somewhat more than a generation ago, started on their way by these methods, were inclined to regard the discovery of the pathogenic agent of some well-known disease as the beginning of the end of its prevalence. To have isolated, recognized and cultivated a bacterium and produced some sort of pathologic changes in an inoculated animal was considered equivalent to half or more of the battle won over the depredations of such organism. For many years these living agents,

but more particularly the unicellular organisms among them, overshadowed all else, and they became synonymous with the causes of disease. Today we know that to have identified the microbic agent of any pathologic process is but the beginning of the solution of the immediate problem, and that it answers but one of a long series of questions. In spite of this somewhat discouraging fact a very remarkable series of discoveries in the biology of disease have originated in this study of microörganisms. I need mention only the bringing to light of bacterial toxins, the discovery of their antitoxins, hypersensitiveness or anaphylaxis, the phenomenon of acquired immunity and the collateral phenomena associated with it, such as the production of agglutinins, precipitins, complement-fixing bodies and, above all, the specificity associated with the action of disease agents and the reaction of the host. Every one of these fundamental discoveries has had a far-reaching influence on the immediate development of the medical sciences. Every one in its way has dominated the thoughts and activities of large groups of investigators, and there has resulted a very unequal, even chaotic, development of our knowledge of the conditions governing disease processes. Throughout this period, dominated in turn by disinfectants, toxins, antitoxins, agglutinins, opsonins, complement-fixation and hypersensitiveness, there is evident some system, some purpose, and that is to find the exact place of living agents in the phenomenon of disease. From the more or less exaggerated point of view held at the start of their dynamic energy in the process there has been a more accurate and more scientific conception of their place as necessary conditions of disease making headway.

It is now evident that the relation of living agents, from the ultramicroscopic forms up to the higher parasites, is different for every agent or at least for every group of biologically related agents. We know now that the depth and extent of their etiologic significance varies from an almost exclusive causation to one of relatively insignificant proportions. For the latter group the environmental and hereditary conditions completely dominate the situation and the particular germ found in one place may be replaced by others in another place. In most disease processes, therefore, the living agents are more or less governed by other factors. This is indicated

by the great variation in the intensity of specific infectious diseases, by their seasonal appearance, by the sudden appearance and disappearance of outbreaks and by the difficulty of maintaining an epizoötic among animals experimentally. Again, it is indicated by the difficulty of inducing disease with pure cultures in species of animals in which the disease occurs spontaneously and in the decline of virulence in artificial cultures.

Now, it may be answered that when we fail to induce disease we do not know how to introduce the agent and where to deposit it. But the how and where are in themselves limitations of the activities of the specific agent. It may also be answered that the microbe with which we try to induce the disease has been attenuated by culture. True, the microbe needs the host to maintain its virulence. This is a significant limitation. Anyone who has been confronted with a disease of unknown etiology, and has in due time found the living agent, knows how little or how much this means when he tries to construct the mechanism of the disease with its aid. In many cases the mystery remains as deep as ever until other necessary conditions have been isolated from the complex of causation.

Perhaps one of the most promising movements to bring into correlation with parasitism the other necessary conditions of disease is the study of epidemiology, not from a statistic but from a biologic viewpoint. To observe that of a given number of exposed animals only a certain percentage contract the disease and only under certain conditions inevitably leads to an inquiry into the causes other than the mere presence of infectious agents.

At this point it might be well to call attention to the necessity for including all invasive living agents however diverse in a study of the factors leading to disease and recovery. In pathology it has been customary to distinguish between parasitic invasion and bacterial infection, the former producing relatively slight disturbances in the host and the latter the acute, often highly fatal epidemic and epizoötic diseases. The distinction is useful, but it can be made to apply only to extremes. Gradations of all shades occur. For the sake of a more exact terminology the distinction between invasion and infection might be made to hinge upon the capacity of parasites to multiply in the host. Thus the number of nematodes

and related worms in the host is not larger than the number of fertile eggs introduced or of those individuals which actively penetrate as larvæ.[1] The formidable power of the infectious agents is due to their capacity for indefinite multiplication in the host. Sexual reproductive stages are not known. Certain protozoa, such as sporozoa, not only pass through sexual stages, but they may also multiply asexually and more or less indefinitely in the host tissues. They may be considered both invasive and infective. It is thus best to class all living invasive organisms as parasites, subject more or less to the same host mechanisms of repression and destruction. At one end of the scale are the highly specialized forms, adapted to one host species or even one race. At the other end are types emerging from the predatory or saprophytic stage and acquiring parasitic habits.

Another concept which we as medical men should have clearly before us is that the phenomena which medical science is chiefly interested in—namely, those of disease—are merely epiphenomena in an evolving parasitism, by-products which tend to lessen and disappear as the parasitism approaches a biologic balance or equilibrium. How rapidly this evolution may progress we have no means of knowing. We do know that among animals epizoötics tend toward a lower level of mortality and morbidity. If we are actually studying by-products it is obvious that to understand them we must first understand the main processes that give rise to the by-products, and that disease is studied most successfully by studying the necessary conditions that give rise to it. If the by-products are in themselves necessary antecedents of other pathologic conditions they would of necessity be included in any study of causation.

A sufficient number of living agents of disease and of parasites has now been studied to permit a tentative classification into highly specialized parasites adapted to and dependent on a given host and those that are more or less predatory, awaiting adaptation provided their organization should permit it. The highly adapted micro-

[1] We must except from this broad statement the nematodes of the genera filaria and trichinella, since their progeny develops within the same host to the larval stage and stops there.

organism which depends upon one host for its existence—as, for example, the still unknown smallpox organism—has through natural selection established between itself and its host a certain balance or equilibrium. This can be defined as a condition of both host and parasite which permits the latter to enter the body, multiply enough and escape so that the arrival of its progeny in another host is assured. On the basis of this relationship we may define four critical phases in the life-cycle of the microbe: (*a*) Its entry into the body and through protecting tissues; (*b*) its transportation to and multiplication in certain tissues; (*c*) its escape from those tissues and from the host as a whole; (*d*) its transfer to another host. Each one of these phases is capable of subdivision into a larger or smaller number of sequences according to the special living agent involved. In the insect-borne diseases the insect acts as transfer agent and as introducer into the blood. The fourth and the first stage merge. In general the fourth stage, or stage of transfer from one host to another, has been greatly modified by civilization. It is therefore necessary in any attempt to formulate the problems of etiology to take into consideration the primitive conditions under which the infectious diseases originally flourished and from which they have come down to us. We do not know whether they originated with man or earlier among his progenitors. But it is safe to take the ground that infectious diseases flourished among the earliest races and that they flourish today among savage and semicivilized peoples as they do among domesticated animals in our midst—that is to say, the infectious agents developed in an environment in which transfer from host to host was direct, immediate and easily brought about. Furthermore, the infusion of susceptible subjects, except during wars and migrations, was slight. These two conditions tended to counteract one another and to bring to an approximate perfection the parasitic habit of the invading organism.

It is this fourth stage of transfer that engaged the entire attention of the early bacteriologists. They created the era of isolation and disinfection by improving diagnostic methods and studying the modes of exit of infectious agents and their resistance in transit. It was tacitly hoped and expected in this great work that infectious diseases could be easily controlled and suppressed by destroying

the agents in the environment of the sick. The science of one generation becomes the practice of the next. Disinfection, isolation and the widening of the danger space between the sick or infected and the well is the chief occupation of modern sanitation. The actual significance of this practice needs to be evaluated from time to time if only in the interests of economy of effort. While it is generally conceded that the movement of the agents of disease should be restrained as much as possible, and while heroic efforts are being made to this effect by health officials, economic forces are driving people together and condensing populations and thereby largely neutralizing the efforts of sanitarians. If any value can be put on this work at present it might be to the effect that it tends to keep individuals from getting an overdose of infection.

When we come to the first phase in the cycle, the entrance of the virus into the body and its penetration through the skin and mucous membranes, our knowledge is, on the whole, neither accurate nor abundant. While the fourth stage has been pretty thoroughly exploited the first is hardly at all known in its details. Each well-established infectious agent will have its own story to tell of this phase. In extenuation of the deficiencies of our exact knowledge it should be stated that the problem is a very difficult one. Micro-organisms leave the body in armies, having multiplied to supply a progeny ample to cover losses in transit. On entering only single individuals or small groups are involved, and unless their morphology is characteristic, like that of the sporozoa and the metazoan parasites, the entry is well beyond the ken of the observer. It can only be got at indirectly. The difficulties of this stage are well illustrated by the prolonged discussions concerning the entry of tubercle bacilli. Much has been done and written in the attempt to clarify this problem. The doctrine of the inhalation of dried bacilli in dust, the droplet infection of fresh sputum and the theory of the alimentary origin of infection have had their day in court. Similarly, the portal of entry of the virus of the eruptive diseases has been the subject of much study and discussion.

The penetration of living agents through the skin and mucous membranes is full of unanswered questions. The resistance of the normal mucous coverings has probably been greatly underestimated.

The effect of injuries in removing this barrier has been similarly underestimated. From a practical standpoint this problem may seem academic, since the mucous coverings, for example, may rarely be free from minor lesions, and such lesions may remain, at best, undetected. But the object of genuine medical science is to get away from the benumbing influence of such considerations. Do typhoid bacilli, for instance, penetrate the normal mucosa or are they dependent on slight lesions? Do protozoa assist them at certain seasons? Do phagocytes ever migrate out under normal conditions and carry bacilli back into the tissues? Might this take place when inflammatory processes are active? Is the entry of certain viruses prepared by other viruses acting only on the epithelium and destroying it or in some other way making the tissues involved more vulnerable? Epithelium-destroying parasites are well known among the sporozoa. It is not improbable that other types of microörganisms, especially those not within the range of the microscope, are specifically adapted as genuine parasites to invade these cells and so prepare the way for the saprophytic predatory types which are readily recognized because easily cultured and therefore regarded as summing up the entire etiology.

After the living agencies have entered the tissues they must run the gauntlet of the blood, lymph and the phagocytic cellular elements to reach those tissues where they multiply. Multiplication is essential, for a large progeny is necessary to cover the losses in transit. This stage of multiplication involves the problem of specific resistance or immunity and susceptibility, and also the practical problem of treatment by therapeutic agents, serums, vaccines and the like. The biologic requisite to be fulfilled by the parasite in this second stage, or stage of multiplication and sexual development within the host, if such a stage exists, is that multiplication must take place in such a way that escape in large numbers from the host after the parasites have assumed a more or less resistant form becomes possible. This is best accomplished when they settle down and multiply near some portal of exit: first, the skin or subcutis; second, the respiratory tract; third, the digestive tract, and fourth, the genital tract. A brief consideration of the localization of the different groups of parasitic agents will show that these various

superficially located tissues are the chief seats of multiplication. Localization in other tissues or organs is rare, and so far as the parasite is concerned, abnormal. If it should happen that a race of Treponema pallidum arose which promptly and exclusively localized in the central nervous system it would die out for want of an exit to another host. The tendency to locate and multiply in tissues of lower vital dignity, *i. e.*, near the surface of the body or the mucous membranes, safeguards the host as well as the parasite.

Among the parasitic invaders of man and the higher animals the metazoan and protozoan parasites have developed relatively perfect and in some cases complicated cycles. The same is probably true of those highly specialized microörganisms which produce the eruptive diseases and of some of the so-called filterable viruses. In fact, all diseases or parasitic states which maintain themselves indefinitely in a host species and are manifestly transmitted from case to case have complete even though not necessarily elaborate cycles.

The tubercle bacillus is frequently referred to as a highly adapted parasite, but its parasitism is crude compared with that of the smallpox organism. It has no well-defined cycle except in phthisis, in which it is inhalation and expectoration. If for the sake of illustration we conceive the primary lesion as leading in every case to a secondary miliary tuberculosis in which there is extensive invasion of the skin with subsequent ulceration, the tubercle bacillus would then be inhaled and after multiplication shed from the skin, in so far approaching the smallpox organism in its cycle. But there is no indication that such a complete cycle ever will be established. On the other hand the leprosy bacillus appears to have in a bungling way reached this stage, for the shedding of leprosy bacilli takes place from the skin and the mucous membranes, notably of the nasal passages.

In certain cases the cycle is limited to a mucous membrane parasitism and the disease is the result of a diversion of the organism into the tissues of the body. The cycle of the typhoid bacillus may be some locus of the digestive tract with incursions into the blood. Whether the invaders perpetuate themselves, *i. e.*, escape again, depends upon ulceration of the lymphoid tissues of the intestines.

The meningococcus resides in the upper respiratory tract. We do not know whether the cocci which enter the central nervous system escape or not. This is an important question, for it is probable that the degree of virulence of the microbe depends upon some contact with the host tissues more intimate than that of a mere saprophytic existence in recesses of the mucous membranes. If such an organism could associate itself with some living or dead factor which assisted its entrance and exit a definite disease might thereby become established and we might expect the disease to rise and fall in epidemic style according as the helping factor is active or not. Contact with and multiplication in living tissues, either cellular or humoral, appears, furthermore, to stabilize the microörganism. If we look over the immunologic and serologic data which have accumulated about the various groups of parasitic agents we find the relationships established by these reactions much more uniform among strictly invasive than among the secondary organisms which depend upon formidable factors to open the way into the system for them. Or to put it another way, there is a much larger number of serologic types among the hangers-on than among those capable of direct invasion.

If the picture I have drawn of the parasitic cycle is fairly accurate it follows that the localization of disease agents or parasites in all tissues and organs, except those from which ready escape to the exterior is possible, is abnormal and unnecessary for the continued existence of the parasite. Hence, typical, characteristic, recurring infectious or parasitic diseases affecting the central nervous system, the ductless glands, the liver and kidneys, the muscular system and the joints do not occur excepting as secondary localizations of diseases involving the more external tissues. If such localization should occur regularly without some superficial localization as well we must look for some source of infection belonging to another species in which the cycle is normal and from which a constant supply of parasites is available even though they fail to escape from the new host. The invasion of the human body by the bovine tubercle bacillus is a familiar case in point. In some infectious diseases several invasions of the same host may be necessary to bring the parasitic cycle to completion, or, viewed from a medical

standpoint, to bring the disease to the clinical level. In bovine tuberculosis, which can be studied anatomically and topographically by killing animals in early stages, the first tubercle bacilli to enter the system usually land directly in regional lymph nodes. To complete the cycle the virus must enter the blood and establish secondary foci in lungs or other tissues from which the bacilli may escape to another host. The cycle is, however, more easily explained by reinfection. The inhaled bacilli lodge now in the lung tissue, due to a changed reaction of the host tissues at the point of entry, and the cycle—inhalation and expectoration, assisted by discharge in the feces through swallowing the sputum—is established.

There are two factors that may modify the normal cycles more or less: One is a relative immunity which may be either natural or acquired. Through immunity cycles may be cut short chiefly in the stage of multiplication and the parasite fail to escape at all or in sufficient numbers to maintain further existence. If for any reason the normal resistance of the host is reduced the parasite may multiply unduly or invade more territory and cause death of the host before the cycle is completed. This condition may be explained by regarding parasitism not as a condition of peace but of armed truce. As soon as one of the two organisms falls below a certain level the other takes the advantage. In case the microörganism gets the advantage it may be fatal for both host and parasite. This latter condition of reduced natural resistance is supplied by civilization either by bringing into the original disease other parasites and greatly complicating what might have been a simple situation, or else by conditions arising from inherited defects, overexertion, abnormal diet, exposures to heat, cold, which are supposed to favor the parasite in its entry and multiplication. These conditions furnish the many modifications of disease types which may be as varied as the number of individuals attacked. It is this increasing complexity of affairs which supports from the scientific side the dictum that the physician is to treat the patient rather than the disease, for there is no longer a type disease to be recognized regularly.

This brings us back to the original subject of etiology. A careful biologic study of the many parasitisms of man and the higher

animals brings out the fact that the highly specialized parasites have no obstacles to their activity except one, and that is immunity either acquired or natural. As a result all host individuals pass through the disease early in life provided opportunity for invasion is given. The highly specialized diseases tend to become in endemic localities children's diseases.

In the case of many other diseases certain non-parasitic factors are necessary to start the disease or to check it, as the case may be. They are part of the mechanism of causation and represent necessary conditions. These necessary conditions may far outweigh the living agents in etiologic significance. The relative importance of the living factors may be so low that their place may be taken by other living agents, or even non-parasitic factors, in the sequence leading to or continuing existing disease processes. This is probably true of certain diseases of intestinal origin. Such diseases are frequently described as due to different microbic agents in different localities because the endemic flora happens to be different.

The relation between the factors of parasitism on the one hand and those of heredity, environment and the like on the other may be briefly summarized as follows:

In the saprophytic or predatory type, representing the so-called septic infections, the other parasitic and non-parasitic factors are of great, even predominating, importance in the production of disease.

In the highly parasitic type they are of little if any importance unless it be hereditary characters brought out by the selective action by the parasites themselves upon the host species through many generations. Many gradations exist between these extremes, and the relation of parasitic to extraparasitic factors is different for the different grades.

Moving parallel with the degree of adaptation and specialization and the development of more nearly perfect cycles by the parasites the mortality drops and the morbidity at first spreads and finally tends to decline in certain types of parasitism, provided always that other types of parasitism do not accidentally enter to modify and complicate the normal course.

In tracing the various living agents through the body of the host

we find that we do not know the details of any parasitism to our satisfaction. These details, of course, include also the non-parasitic factors or conditions essentially favoring or hindering the parasite in its sojourn in and its journey through the host tissues. It is perhaps needless to refer at length to the conditions which control the acquisition of such knowledge. The hope of finding some preventive or cure dictates the course of many workers. When something approaching this has been found the etiologic significance of all the other factors bearing on the disease falls below the horizon for the time being. The importance of continuing the study of the disease persists, however, even for practical reasons, for the remedy or preventive may not prove to have the success anticipated. To induce men to fill the gaps of our knowledge seems quite as important as the pioneering for entirely new vistas or outlooks. Great discoveries are, as a rule, half-truths that must be brought into line by patient after-research. The filling of gaps may be necessary to stage the next great discovery.

There is beyond the mere knowledge that gaps exist the difficulties of the problems involved to be considered. Are we prepared to solve them directly or must we rely on indirect approaches and on the solution of analogous problems to satisfy our etiologic sense? Added to these difficulties, inherent in some phases of the parasitic cycle, notably that phase which takes place within the host, is the fact that the various disease agents attacking the same species, man for instance, are so different from one another that we may safely consider them surviving types. It would seem that where two or more parasites follow the same route and multiply in the same tissues a certain competition tends to eliminate one or the other. If two closely related types exist they rarely multiply in the same host at the same time. Such competitive elimination would leave a divergent assortment of parasitic organisms and resulting diseases, none of which would be an exact copy of the other. In this case the working out of one cycle does not necessarily enable us to predict what another would be. They must each and all be studied individually.

In this dilemma we may gain assistance from a study of parasitisms in those remote and isolated regions which have not yet been

seeded by the white man's diseases, where the prevailing maladies may still be "pure lines" rather than mixtures and combinations. Another source of material are the many characteristic parasitisms of animal life, notably of the mammals and birds. Comparative pathology may furnish us with that information which experimental pathology finds it impossible to produce. Taking all the diseased and abnormal states due to living agents in man and the higher animals together a series may be established which fills in many gaps and which may furnish the suggestions and clues needed to bring about a better insight into the dynamic relations between host and parasite. Only through the coöperation of comparative and experimental methods may we hope to gain enough general underlying concepts to explore with some show of rationality new diseases successfully. Since science is valued in proportion to its capacity to predict successfully certain events, medical science will be judged by the way it takes hold of a new phenomenon to determine its etiologic antecedence. If in the course of its development it has failed to take cognisance of factors necessary to build the science into a consistent whole it should retrace its steps and make up the deficiency.

Parallel with the continued analysis of phenomena there should be another process going on to simplify the complexity resulting from the former and to bring the results of scientific inquiry more or less within the reach of every-day life. What is needed is a synthesis of the many data resulting from the analytic study of phenomena. Perhaps I can make myself clearer by using as an illustration some recent investigations: If we examine the various diseases in which the virus is conveyed by insects and arachnids we shall find that many of the data pertaining to the dissemination of the virus had been accurately worked out before the mode of transmission was discovered. There was lacking, however, a something to harmonize and coördinate them. When the insect carrier was defined these various discrete apparently unrelated data fell into line. Here was a synthesis which not only substantiated older observations but it enabled the scientist to use the deductive method to develop new inquiries and thereby lift the subject up to a higher level for further analysis. For some years we had known that a certain disease of

young turkeys, due to the invasion of the tissues through the intestinal tract by a protozoan parasite, could be prevented by raising these birds away from older turkeys and common poultry and on soil uncontaminated by them. The explanation came through the discovery that a common worm of these species was needed to injure the mucous membrane and thereby open the way for the protozoan parasite. The nematode also accounted for certain disturbances in the application of the above rules in the rearing of these birds. It synthesized, in other words, the accumulated data.

With the aid of these illustrations it is possible to understand, at least in part, what must have been the effect of the rapid discovery of various living agents in the 80's of the last century on the medical mind of the period. Many apparently unrelated data suddenly moved into line and assumed definite relations to one another. The discoveries pertaining to acquired resistance to disease involving the action of antitoxins, agglutinins, precipitins and the like have not had as yet the desired effect of synthesizing the conception of immunity, because they may be accessories rather than essential factors, all grouped around some more fundamental, unifying, still undefined phenomenon.

THE ROLE OF THE PARASITE IN INFECTIOUS DISEASES[1]

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The discovery of the existence of microscopic forms of animal and plant life, and the proof that these minute organisms arise by continuous descent from other like organisms, is, from an historical standpoint, a comparatively recent addition to the knowledge of natural phenomena. Since the establishment of these two fundamental facts the time and attention of many workers in the field of science has been devoted almost exclusively to an effort to comprehend the varieties, the natural history and the biologic activities of these minute bacterial forms. The field has been a fruitful one for investigation, and there has been a rapid extension of our understanding of the profound influence that bacteria have on life-processes. This influence may be beneficent, as in the instance of soil bacteria, or it may be malevolent, as in the case of the parasitic and disease-producing organisms of animals and plants. Man's interest lies largely in the activities of the latter group, for it is with them that he is most frequently brought into competition, and for his own defence he has been forced to cultivate intensively all fields of study which hold the promise of increased knowledge and power in the struggle. From this necessary and practical investigation has come much of the information which we now possess concerning the pathogenic types of bacteria which comprise the group of particular interest to students of infectious diseases.

Infection is generally defined as the penetration of living tissues

[1] From the Biological Division of the Medical Clinic of the Johns Hopkins University.

by small microscopic forms of life which possess the capacity to multiply in such tissues. This conception of the etiology of certain diseases is an ancient one, but only in recent years have the facts been laboriously accumulated which have demonstrated the truth of this idea. We now know, largely from the investigations of Pasteur and Koch, that many diseases are caused by invasion of the body by bacteria which in most instances bear a specific relationship to the resulting pathologic process. A consideration of the incidence of infectious diseases in the world today shows them to exist in all known portions of the globe—some with a well-defined regional distribution and others more or less universally distributed. Whatever the spatial separation may be, a striking similarity in the biologic aspect of different instances of the same infection is manifested throughout the world. The general trend seems to be in the direction of some relatively stable state, though change in the character of certain specific infectious diseases has probably occurred, and that still further variation may develop in the future is not unlikely. Few diseases that we know of have spontaneously disappeared from life, and of even greater interest is the fact that no definitely new infectious disease is known to have arisen or probably ever will arise. It is, then, a matter of great importance to examine the series of phenomena by means of which such an equilibrium has been established and is maintained, inasmuch as the possibility exists that once all the facts are known, this apparently static condition of affairs may be disturbed and the war against infection may more and more be carried on in a manner favorable to conservation of human life.

One of the first questions with which the mind of the investigator in bacteriology is confronted concerns itself with the mechanism by which certain bacteria have completely adapted themselves to the parasitic habit, and continue to produce an unending succession of instances of disease, whereas the great majority of species are content to lead a harmless saprophytic existence. Have some species from the very beginning possessed the necessary invasive qualities requiring only the opportunity of the right sort of contact to start them off on a career of parasitism? or has the chance come only to the occasional strain, possibly in an accidental manner, but

with each succeeding opportunity attended by a gradual perfection of the weapons of offence? The origins of infection are, of course, lost in obscurity, but even today we are constantly meeting micro-organisms which vary greatly in the continuity of their parasitic phase and the facility with which they are capable of entering into this cycle of their existence. For example, Bacillus tetani must be considered as a purely accidental parasite needing a particular and unusual group of favoring conditions to permit it to establish itself successfully in the living host. It is indeed likely that such a strain having gained entry into the animal body produces but a single instance of disease, and then ends forever its career as a parasite. On the other hand, Treponema pallidum has advanced to such an exquisite form of parasitism that it invades living tissue with the greatest ease, maintains itself there for long periods of time without undergoing serious injury and cannot sustain life at all if removed for any length of time from its acquired environment. Has treponema passed through a stage of discontinuous parasitism somewhat analogous to the occasional and accidental parasitism of Bacillus tetani observed today, and has it gradually perfected its invasive qualities to the disadvantage and final loss of its saprophytic powers? If this is so, why do we not see the occasional parasites of today rising in the scale of parasitism to the degree manifested by so many disease-producing organisms at the present time? Can it be that the conditions for the origin of new permanent parasites have become unfavorable and that this change is due to a developing unsuitability of the soil?

That all bacteria, including even the most parasitic species at some period of their existence, have been able to live free in Nature and to survive during such a saprophytic phase is not unlikely. Indeed, the possibility exists that saprophytism represents the primary state of these microörganisms. If such should be the case, then the opportunity must early have been offered for implantation on the exposed skin and mucous surfaces of living beings, and the suitability of such an environment is demonstrated by the large number of species that are constantly found today in these localities. The facility with which such implantation occurs is shown by the rapidity with which the intestines of newborn infants and animals

become infested with colon bacillus. That the majority of such organisms are fortunately non-pathogenic is a matter of common knowledge. However, if a careful analysis of such groups of bacteria is made the interesting circumstance is encountered that in general each group has at least one pathogenic representative which, in its gross morphologic and cultural characteristics, and in some instances even in its finer biologic attributes, closely resembles the non-pathogens of the group. Thus staphylococcus and streptococcus are found quite constantly on the skin and mucous membranes of human beings, and for each there is a highly pathogenic and virulent analogue which is responsible for numerous severe and often fatal infections in man. Among the many closely related types of Gram-negative cocci of the nasopharyngeal mucous membrane there exists the meningococcus which frequently causes a highly fatal form of epidemic disease. Examples of such other groups are the pneumococcus and the colon typhoid dysentery series, and if the entire field were accurately studied, no doubt many other instances of this phenomenon would reveal themselves. In some of these groups the pathogenic member is readily distinguished by cultural and biochemic reactions, whereas in others this is only accomplished with difficulty by means of reactions of such extraordinary specificity as those of immunity. The significance of this state of affairs as concerns parasitism is difficult to interpret. The possibility exists that in early times many strains were occasionally pathogenic, but that a developing resistance of the host has brought about selection of an especially equipped few, and that, as the pathogens of any given group have developed their invasive powers to a high degree, the resistance of the host has risen to withstand this danger, and that this very phenomenon has operated as a check to the less infective races. Whatever mechanism is the real one, certain it is that a separation into pathogens and non-pathogens has occurred, and that this differentiation is a fairly fixed one, and that the development of new pathogenic species is not observed.

In recent years there have come into use for purposes of bacterial classification various immunological methods, such as protection, agglutination, precipitin and absorption of antibody reactions. By means of these reactions finer and more reliable distinctions between

different types of the same bacterial species can be determined than by the use of the more common cultural and biochemic methods. Though these procedures are receiving an ever-widening application in bacteriology, up to the present time no single group of bacteria has been completely worked out so that the exact position of every strain can be accurately determined. In spite of the incompleteness of the study, however, certain facts have been brought to light which possibly are of some significance in the study of parasites. An example of this type of bacterial analysis is furnished by investigation of the pneumococcus group, in the understanding of which a considerable degree of progress has been made. The pneumococci constitute a group of organisms which resemble each other very closely morphologically and in their staining properties. The colony forms and the character of their growth on artificial media is identical with a single exception, that of the so-called pneumococcus mucosus. When examined biochemically by means of various test sugars their fermentation reactions are found to be similar, and in general the pathologic processes which they induce in animals are the same. From all the evidence which can be obtained from such standards of comparison the conclusion must be drawn that each strain of pneumococcus exactly resembles every other strain. However, when such a series of similar strains is examined by means of certain immunologic reactions it is found that sharply differentiated types exist. Each individual of a given type is identical in all its immune reactions with every other member, but the types themselves are readily distinguishable. Three such fixed types have been established and are known to be common in widely separated parts of the world. In one limited locality—South Africa—other fixed types which do not occur elsewhere have been observed. These fixed types of pneumococcus manifest their specific characters with great regularity, and over fairly long periods of time, and in addition strains of exactly similar type are continually being isolated from their natural environment. In addition to these fixed types of pneumococcus there exists a great mass of strains that are dissimilar in their immune reactions, each strain from the standpoint of immunity constituting a biologic unit. The importance of this state of affairs for the study of parasitism lies in the fact

that the pathogens of the group in general fall into the homogeneous types of pneumococcus, whereas the less pathogenic and the more or less harmless strains of the normal mouth secretions are heterogeneous in character. Such a separation is, of course, not absolute, and there is some overlapping in both directions. Analogous conditions are found among other groups of disease-producing bacteria. Gordon[1] has shown that 96 per cent of the epidemic strains of meningococcus can be placed in four separate types and others have denied the specificity of the types of meningococcus because of the heterogeneity that is exhibited by strains isolated from normal individuals in communities in which cerebrospinal fever has not been epidemic. As a further illustration of this situation, one has only to call to mind the high degree of specificity manifested by the typhoid and paratyphoid bacilli in comparison with the great diversity from an immunologic standpoint shown by different strains of Bacillus coli. In general, then, the more highly pathogenic a microörganism the more likely it is to be comprised within a type exhibiting uniformly specific immunologic relationships; whereas the more saprophytic organisms show the greatest possible degree of diversity—in fact, immunologic unity is becoming more and more a criterion for the determination of disease-producing capacity. There are two possible interpretations of this interesting phenomen: Highly pathogenic races of bacteria may either be pure lines from the standpoint of descent, or, with the acquisition and perfecting of the parasitic habit, they may develop, by adaptation to a fixed environment, a high degree of immunologic uniformity.

I realize that much more study is required—in fact a beginning has hardly been made—before we can discuss the subject of parasitism with any assurance of its proper understanding. However, certain observations may furnish suggestions for progress in the future, though the explanation that is given them is in no way to be regarded as final. Thus the disease lobar pneumonia among the black workmen in the mines of South Africa exhibits certain characteristics from the point of view of bacteriology which possibly are of some significance in the understanding of the development

[1] Jour. Hyg., 1918, xvii, 290.

of pathogenicity among certain types of pneumococcus. Laborers for the Rand diamond and gold mines were formerly recruited to a large extent from the more tropical regions of Africa, localities in which severe infections of the respiratory tract are uncommon. These men were finally gathered together in a huge compound at Johannisberg in numbers as many as twenty thousand at one time. Within a few days from the beginning of such a concentration, lobar pneumonia of pneumococcus origin made its appearance in the compound, and the incidence and mortality of this disease quickly rose to a very high point. This high pneumonia rate continued even after the black boys were sent to their ultimate stations in the mines. Lister[1] has made a study of the bacteriology of these instances of lobar pneumonia. He has found that the number of fixed types of pneumococcus causing pneumonia among the blacks is greater than that observed either in South Africa or throughout the rest of the world among the white races. In fact, the dominant type in the pneumonia of the blacks does not exist at all as a fixed and frequent type among the whites. In other words, certain new and well-characterized races of disease-producing pneumococci seem to have obtained a foothold in this highly susceptible population. The only interpretation of this phenomenon that I can offer is that certain strains of pneumococcus which produce only occasional instances of pneumonia among the more or less resistant white peoples, and then because of their low degree of infectivity end for the time being their career as pathogens, when communicated to the black find the ground more suitable and are readily communicated in a virulent state from one individual to another. Thus they quickly perfect their disease-producing powers and soon come to occupy the field to the exclusion of other strains. This high pneumonia rate has been observed in other parts of the world under similar conditions, but has never received such careful bacteriologic analysis. The one common factor which has been present in all such epidemic outbreaks of pneumonia has been the bringing of a highly susceptible mass of material into intimate contact with a presumably more resistant race in which pneumonia has been endemic in incidence.

[1] Publication of the South African Institute for Medical Research, 1914, W. G. Horter & Co.

It is interesting to speculate whether the exposing of a large number of susceptible individuals to an unaccustomed form of infection does not create conditions favorable to the acquisition of virulence by certain relatively non-pathogenic races of the organism concerned. Observations of this nature seem to indicate that a high degree of racial susceptibility may furnish environmental conditions which are suitable to the development of pathogenicity by bacteria. It is possible that in early times many organisms possessed latent invasive qualities, and that during the period of high racial susceptibility certain strains representative of different bacterial species quickly availed themselves of the opportunity offered. The resistance of the host species has probably risen to meet this danger and the law of selection has become operative. Only the types of organism best equipped offensively and defensively have survived as the pathogens of the species, and the less adaptable members of the group, having missed the original opportunity, fall behind and out of the race. Only with the shifting of populations are the original conditions in any way repeated and a limited opportunity again offered for the acquisition of disease-producing powers.

However, certain microörganisms have passed from a saprophytic phase to one of more or less continuous parasitism; it is not unlikely that this transition has been attended by the acquisition of new and distinctive qualities both of structure and function, which are transmitted from generation to generation. The accurate determination of such possible changes is seen to be one of great difficulty when we consider that virulence or pathogenicity does not lend itself to direct measurement, but must always be estimated in relation to some other living organism. Infection has come to be looked upon as the manifestation of a struggle between two opposing forces—the life-force of the invading parasite and a similar force of the invaded host. The greater the opposition the more severe is the struggle and the less likely the establishment of an equilibrium which is advantageous to each participant. This changing warfare with its ever-varying adaptation of the offensive and defensive mechanism of the parasite to meet the changing conditions imposed upon it by the host is the problem with which

we are confronted in infectious disease today. The reaction of the host will be considered in another paper, and Theobald Smith[1] has previously discussed in detail those factors which are serviceable to the maintenance of the life-cycle of the parasite, so that it may serve to mention but briefly here some of the more important of the latter.

Virulence may be described as that quality of bacteria by means of which they invade and multiply in living tissues. As yet we possess no general method for determining the degree of virulence of any pathogenic organism for man. It is true that many bacteria which cause human disease are also able to bring about infection in the lower animals, and, to a certain degree at least, virulence can be measured by the use of such experimental animals. The results obtained in this way, however, are very irregular, and numerous instances exist in which the pathogenicity of certain microörganisms is exhibited in only a very limited number of species of animals. In fact, in some cases passage of an organism through a species of animal other than that which it naturally infects, results in marked attenuation or even complete loss of virulence for the original species. Some attempts have been made to correlate growth-energy with invasive power, but so far no distinctive differences have been observed between parasitic and non-parasitic races. Some disease-producing bacteria, such as Bacillus diphtheria and Bacillus tetani, are known to possess and use weapons of a definitely offensive character. By means of specific poisons they destroy the defence mechanism of the host and thus obtain a local foothold. Such organisms are, however, not highly invasive, rarely spreading to any extent from the site of inoculation, so that toxin-production, as Theobald Smith has stated, can hardly be regarded as a permanently useful quality in the struggle upward to a high plane of invasiveness. Of more use to bacteria for purposes of invasion is probably the capacity to produce hemotoxins and the so-called aggressins of Bail,[2] types of activity which are designed to paralyze the more general defensive mechanism of the host, and thus reduce not only

[1] Am. Med., 1904, viii, 711.
[2] Arch. f. Hyg., 1905, lii, 272.

the local resistance but destroy throughout the body the specific mechanism of defence. The aggressins are of special interest in this connection. Bail has shown that exudates caused, for instance, by Bacillus anthracis, when free from living anthrax bacilli possess the capacity to increase the invasiveness of this organism. For example, a non-lethal dose of anthrax bacilli when injected into the living animal in combination with a small amount of such cell-free exudate rapidly produces a fatal infection, whereas control animals receiving the organisms alone continue to live. This phenomenon has been explained by the statement that there are present in the exudate substances derived from dead and disintegrating anthrax bacilli which neutralize the natural antibodies and thus favor infection. Such an explanation is, however, hardly adequate, inasmuch as we know that the pneumococcus during life, and at a time when no cell-destruction is occurring, secretes a large amount of material, the source of which is the living cell, and which possesses the capacity to neutralize and throw out of solution the specific pneumococcus antibodies. In fact, this substance is present in easily demonstrable quantities in the circulating blood and urine of individuals suffering from severe pneumococcus infections. That other bacteria, especially the capsule-bearing varieties, give off during life a similar substance is not unlikely.

As bacteria advance in the scale of parasitism, as Theobald Smith has pointed out, they seem to lose their offensive weapons and to depend for continued existence upon the perfection of their defensive mechanism. Thus such well-adapted organisms as Bacillus tuberculosis and Treponema pallidum exhibit a comparatively slow destructive action within the body and may resist dissolution for long periods of time, the one because of its highly resistant membrane and supporting structure and the other because it stimulates such slight general reaction that antibody formation seems not to occur. Other organisms, especially when in a highly virulent state, seem completely insusceptible to the phagocytic activity of the leukocytes, and are thus enabled to survive under conditions in which less virulent races are quickly ingested and killed within the bodies of the white cells of the blood. Ability to form capsules is also an

important defensive mechanism of bacteria. Bordet[1] has observed that in experimental infection of rabbits with streptococcus only those organisms survive and multiply which possess the capacity to develop capsules, and Ascoli[2] has reported a similar observation in regard to anthrax bacilli. Capsule formation and invasive power, however, are not necessarily parallel, for Friedländer's bacillus is distinctive because of its well-developed capsules, and yet possesses but little power of independent invasion of the tissues.

That passage of pathogenic microörganisms through a series of susceptible animals enhances the virulence of such organisms is well known. A similar effect probably results from the rapid transfer of bacteria from man to man in the natural course of an infectious disease. If this is the case we may well ask, Why does not the mortality from infectious diseases continually increase instead of tending to diminish, as seems to be actually the case. Theobald Smith explains this phenomenon by pointing out that a parasite in order to survive must not only be able to invade the body but must also be provided with a means of escape in order that it may bring about the infection of new individuals. If an organism becomes so virulent that it enters the blood without hindrance and rapidly brings about the death of the host, then its chances of escape are lessened by the short duration of the infection and by the diminished likelihood of the production of open lesions near the body surfaces, from which an easy means of exit is provided. Thus the most virulent parasites tend to destroy themselves by the very fact that they produce death rapidly, and in the long run selection favors those less virulent strains whose destructive activity manifests itself over a longer period of time.

The fact that the cultivation of pathogenic microörganisms outside the body on artificial media results in most instances in gradual loss of virulence needs no discussion. Some interest, however, is attached to the fact that bacteria undergo alterations in virulence when grown in contact with a homologous immune serum. The statement has been made that the cultivation of Bacillus typhosus in antityphoid serum increases the virulence of this organism.

[1] Studies in Immunity, J. Bordet and his collaborators, collected and translated by F. P. Gay. John Wiley & Sons, New York, 1909.

[2] Centralbl. f. Bakteriol., 1 te. Abt., orig., 1908, xlvi, 178.

The criteria for acquisition of virulence by Bacillus typhosus are not satisfactory, however, and much significance is not to be attached to this observation. On the other hand, Stryker[1] has shown that when highly virulent pneumococci are grown for several generations in low dilutions of homologous antipneumococcus serum a rapid and complete loss of virulence takes place. No measurable diminution, however, occurs when the same strain was contemporaneously carried for the same number of generations in normal horse serum. There is no question as to the actual loss of virulence, since the titrations were carried out in a highly susceptible animal over a wide range of dosage. Too much significance could hardly be given to this observation if it means that specific immunity exerts an influence in the direction of depression of bacterial virulence. We have been accustomed to think that pathogenic bacteria increase in virulence in order to match the rising resistance of the host.

The teaching of Koch has led us to seek in each infectious disease a single etiologic agent, and adherence to this conception has been responsible for the ever-increasing clarification of our knowledge concerning infectious processes. There are, however, certain diseases, especially among the lower animals, in which two microörganisms are found in practically constant association. In hog cholera, for instance, the hog cholera bacillus is always present in connection with a filter-passing organism, and was formerly supposed to be the sole cause of the disease. It is now known, however, that the filtrable virus is the primary agent and can produce the disorder in experimental animals in the absence of the secondary agent, Bacillus choleræ suis. The occurrence of secondary invasion in human disease is a very common occurrence, and the high mortality of many infections is directly dependent upon the ease with which secondary invaders follow in the wake of the primarily infecting microörganism.

That such secondary invasion is of considerable significance to the phenomenon of parasitism there can be but little doubt, and it is a matter of great importance in the control of infectious disease to determine the number and variety of bacteria at present considered

[1] Jour. Exp. Med., 1916, xxiv, 49.

intrinsically pathogenic which in reality require that the ground be prepared by some primary virus before they are able to develop effectively their pathogenic qualities. An interesting example of the possible succession of events in an instance of this kind has recently been furnished by the widespread epidemic of streptococcus bronchopneumonia which occurred in the spring of 1918. The concentration of troops in the summer and fall of 1917 was quickly followed by numerous outbreaks of measles, especially in southern and southwestern camps, where much highly susceptible material was present. Measles, a disease of low mortality in adults, as a rule, soon came to manifest a very high mortality. Investigation of the cause of this greatly increased death-rate showed that it was almost entirely due to a secondary bronchopneumonia, and that by far the most important etiologic agent in this complicating pneumonia was Streptococcus hemolyticus. Because of the facility with which transference of the organism could take place the distribution of streptococcus became almost universal in measles wards, and in some camps as many as 80 per cent of samples of the entire healthy population were found to be carriers. Before long Streptococcus hemolyticus became common as a secondary invader in lobar pneumonia as well. This was followed by a most interesting development. Numerous instances of primary streptococcus bronchopneumonia were observed both among soldiers and over a wide area in the civilian population, cases in which no predisposing factors of an infectious nature were discernible. In normal times primary streptococcus bronchopneumonia is a rare occurrence in healthy young adults. The most satisfactory explanation of this circumstance would seem to be that streptococcus, an organism that is essentially a secondary invader, obtained a foothold first in patients suffering from measles and lobar pneumonia, and by rapid passage from individual to individual so increased its virulence that it quickly became able to initiate serious infection without the assistance of other microörganisms. A somewhat similar phenomenon accompanied the recent pandemic of influenza which provided suitable opportunities for a great increase in the incidence of secondary invasion of the respiratory tract. The complete significance of such a sequence of events in the development of

increased pathogenicity by organisms of primarily low invasive powers is not altogether understood at the present time. Such an occurrence manifests itself most frequently in connection with diseases of the respiratory tract, and it is interesting to speculate whether it may not be more common than we at present realize. Every year in temperate climates there occur several pandemics of catarrhal inflammation of the mucous membranes of the upper respiratory tract. It is well known that increased incidence of lobar pneumonia is commonly associated with an outbreak of common cold. Is it possible that so important a pathogenic organism as pneumococcus, usually considered a primary invader of the respiratory tract, needs some assistance each year at the beginning of its disease-producing career, but that it rapidly passes through the phase of secondary invasion, and with the accession of virulence quickly acquires the independence necessary for the production of primary disease? Study of the common cold gives rise to the impression that this infection is all too lightly regarded and that it may be the foundation upon which are based the activities of some of the more dangerous pathogens gaining entrance to the body through the respiratory mucous membranes.

It has been customary to divide infectious diseases into two groups: Those which manifest principally sporadic incidence, the so-called endemic diseases, and those which give rise to definite outbreaks at varying intervals of time, infections which are epidemic in character. This distinction is somewhat arbitrary, and any infectious disease under a proper set of conditions may reach epidemic proportions. In fact, certain of the so-called endemic diseases, such as lobar pneumonia, assume a limited epidemic character at certain seasons of the year, so that in all probability no essential difference exists between these two types of infectious disease. The morbidity rate at any one time may be dependent upon certain relationships of parasite to host which are not necessarily constant, and any variation in this relationship may result in great changes in incidence. The factors which enter into such changes in equilibrium are not known at the present time. However, attention has been focussed upon this problem in recent years, and while study of an infection with a view to its solution during

the sporadic or interepidemic phase is too difficult to be undertaken with profit, on the other hand investigation of the epidemic phase is beginning to yield results of considerable interest.

The spread of bacterial infection is a question of too wide an aspect to be discussed in detail in this paper, and I can simply draw attention to certain studies which seem to indicate that variation of the parasite may be one of the most significant factors in the changing incidence of an infectious disease. Topley[1] has investigated this question experimentally and has come to some interesting conclusions. In considering the rise and fall of an epidemic wave with a view to finding some explanation of its origin and rise he says there are at least three explanations: "There may be an increase in the power of the parasite to produce disease, a decrease in the resistance of the host or some alteration in the surrounding circumstances which favors the transmission of parasites from case to case without any alteration in the pathogenicity of the one or in the resistance of the other. The third of these hypotheses may be disregarded. That alterations in environment may be the determining course in initiating an outbreak of bacterial disease is probable enough, but they will almost certainly act through the variations which they bring about in the other two factors. Again, while we may well believe a lowered resistance of a certain number of the host species to be an important factor in the initiation of the process, yet we cannot believe that it is the whole story. The widespread ravages of many epidemics would seem altogether to preclude such an explanation. We seem forced, therefore, to the conclusion that an increase in the pathogenicity of the specific parasite is an essential factor in the rise of epidemics.

"In considering the decline of the epidemic we are faced with three similar possible factors: An alteration in the parasite, now in the direction of lessened pathogenicity, in the host species, in the direction of heightened resistance or in the environmental conditions resulting in lessened opportunities for the transference of the parasite. There is, indeed, in the case of any considerable epidemic, a fourth possibility—the elimination of all susceptible individuals

[1] Lancet, 1919, ii, 1.

by an attack of the disease. This explanation has unanimously been rejected by epidemiologists on various grounds. Brownlee has shown that it is entirely incompatible with the observed symmetry of the epidemic curve and that a progressive variation in the biologic activities of the parasite seems to offer the most satisfactory explanation of the facts observed. While there seems little room for doubt that increased pathogenicity of the parasite must play an essential part in the rise of the curve of disease, it is much more difficult to decide on the relative importance of variations in the powers of parasite and host in bringing about its decline."

With the progress of studies of this subject, however, the evidence seems to point more and more strongly to an essential change in the character of the parasite as the principal cause of the rise and fall of the epidemic wave. Danysz[1] and Topley,[2] studying the infection of rodents with Bacillus danysz, have both shown that as the virus passes through successive series of animals there is for a relatively short period of time an increase in its infectiveness; but that this is followed by a decline in this quality, and that after a limited number of transfers the organism loses this increased infectivity, falling to its original level or even lower. Certain experiments were then carried out to determine whether the spread of infection by contact showed features which would indicate that such a variation in the character of the parasite is an important factor in the spread of the disease. For this purpose small epidemics were started by placing normal mice in contact with others infected by feeding, and small numbers of mice were subsequently added at irregular intervals to test the risk of developing a fatal infection incurred by entrance to the cage at any given time. The result seemed to indicate that those normal mice added during the early stage of such small epidemics had little chance of survival, while of those which were added during the later stages a large part remained alive and apparently well, although at the time of their introduction to the cage, deaths were still occurring. Experiments by both Danysz[3] and Topley[4] indicate that if susceptible mice be continuously added to an infected population the spread of infection

[1] Ann. de l'Inst. Pasteur, 1900, xiv, 193.
[2] Loc. cit.
[3] Loc. cit.
[4] Loc. cit.

will continue over a long period of time, and that there is no evidence that this period has a limit. The significance of the continued addition of fresh susceptible material to the infected population is shown by the observation that the rate of extinction of a population among which infection is rapidly spreading will be far less rapid if they are kept isolated than if further susceptible individuals continuously gain access to them. A proportion of the infected population which would have survived indefinitely under the former circumstances will die under the latter. The studies of Flexner and Amoss[1] seem to indicate, on the other hand, that in spite of the repeated addition of numbers of susceptible mice the mortality in late waves of infection due to Bacillus typhimurium tends to fall to a low level and possibly to disappear. My own observations lead me to believe that this same organism may be maintained at a high and practically constant level of infectivity by continuous passage through susceptible animals. The final interpretation of all these experiments cannot as yet be given. However, there seem to be two points of outstanding interest: (*a*) That an infection started in a fixed population tends to disappear spontaneously, leaving a variable percentage of survivors that are not of necessity resistant to the causative virus at certain phases of the epidemic cycle, and (*b*) that the continuous addition of new susceptible animals prolongs possibly indefinitely the period of infectivity, and that during this period not only the freshly added animals but also numbers of the original population become infected and die. The suggestion might be made that in general the trend is in the direction of the establishment of a balance between resistance and infectivity in a fixed population at a level unfavorable to the parasite and that the introduction of new infectible material gives the parasite an opportunity to refresh its invasive powers, permitting the development of a second wave of infection affecting both survivors and new animals, and eventually resulting in the reëstablishment of an equilibrium at a different level.

In the effort to discover what biologic laws, if any, are operative in the phenomenon of parasitism the student is confronted with

[1] Reported before the Association of American Physicians at the current meeting (1921).

many difficulties, some of which at present appear almost insuperable. Experiments which might lead to the development of adequate generalization appear to be impossible of construction. The transformation of hypothetical free-living species of bacteria into parasitic species is lost in obscurity, and inasmuch as no new infectious diseases seem to arise, opportunity has never been afforded for the observation of a single such phenomenon. On the other hand, study of the influence of one disease-producing organism upon the invasive powers of another has yielded valuable information and may finally answer the important question as to whether certain bacteria are essentially secondary invaders and lack the capacity to initiate disease unaided. Furthermore, comparison of individual characteristics of pathogenic microörganisms with those of nonpathogenic has brought to light many specific properties upon which the quality of invasiveness may be dependent. The kind of investigation giving most promise of the perfection of our understanding of the present status of infectious disease seems to be the study of experimental epidemics in animals. It is most likely that by this method much light may be thrown upon those underlying factors of virulence of the parasite and resistance of the host, the balance between which seems to determine the ultimate course of any single outbreak of infectious disease. These factors when operative over long periods of time may serve to change the character of any infection both in regard to morbidity and mortality. Only by investigation of the above special and accessible phases of the problem may enough related facts eventually be discovered to permit some logical and presumably useful generalization concerning the phenomenon of parasitism as a whole.

STUDIES ON THE TUBERCULIN REACTION AND ON SPECIFIC HYPERSENSITIVENESS IN BACTERIAL INFECTION

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For a number of years we have been interested in defining, if possible, the role which anaphylactic sensitization to bacterial cell constituents might play in the symptomatology and pathology of infectious diseases. We have studied this problem from a number of different points of view, and some years ago published, with J. T. Parker, a series of observations on the sensitization of guinea-pigs with typhoid bacilli in which we made use of the isolated uterus method of Dale.

It has been quite apparent to all workers in bacterial anaphylaxis that the sensitization of animals to the cell materials of bacteria is much more difficult than is analogous sensitization with sera, egg albumen, etc., and it has usually been assumed, and we believe correctly, that this was due to the relatively small amount of coagulable protein in the bacterial cell.

In thinking about the problems of bacterial hypersensitiveness since that time it has seemed to us of fundamental importance to obtain, if possible, a clear understanding of skin reactions such as the tuberculin, typhoidin, mallein, etc., tests which represent more or less specific forms of hypersensitiveness in infected animals and man, but, at the same time, differ in a number of important and perhaps fundamental aspects from phenomena generally associated with true anaphylaxis.[1]

[1] By true anaphylaxis we mean the reaction of protein hypersensitiveness, in which it is now generally agreed an antigen-antibody union is involved in which the predominant role is played by the sessile antibodies and which can be passively and specifically transferred to normal animals with the serum of other animals so long as this serum contains antibodies. We do not wish to take up space with this definition but refer the reader to the recent and excellent summaries of Doerr, Coca and Wells given among the references. Although differing somewhat in nomenclature and in classification, the three summaries lay down fundamental principles which are essentially alike and with which we agree.

The work reported in this communication is based primarily upon an attempt to elucidate these forms of skin hypersensitiveness to bacterial products, and to gain, through the study of these reactions, some light on bacterial hypersensitiveness in general.

I. The Two Different Types of Skin Reaction. A basic fact in regard to skin reactiveness is the occurrence in man and in guinea-pigs of two fundamentally different types of such reactions, both of them to a considerable degree specific, yet differing both in the nature of the completed reactions and in the time factors concerned in their development. We specify guinea-pigs and man because in rabbits the distinction is not so clear, a matter to which further reference will be made below. The two forms to which we refer are:

1. The intradermal reaction, which appears in from two or three to fifteen minutes after injection of the antigen. It expresses itself in the development of a growing wheal, often surrounded by a red areola of variable size. This reaction may last from one-half hour to one or two hours, and fades again, usually without leaving any profound injury of the tissues. This is the reaction obtained with such substances as horse serum, and has been extensively used in man to determine whether or not the particular individual was sensitive to horse serum when therapeutic injections were intended.

2. The other type of skin reaction is one in which there is no immediate effect but in which within four, five or more hours a swelling becomes apparent which in the course of twelve to twenty-four hours results in a swollen, edematous area of varying intensity, often with a central necrotic spot and occasionally hemorrhage. This reaction may not reach its highest development until about forty-eight hours after the injection, and is accompanied by distinct signs of inflammation and some cell death.

As we shall see the two reactions may occur in the same animal, after one and the same injection, but in guinea-pigs can usually be differentiated, since one begins to fade before the other appears.

The classic examples of such skin reactions are the tuberculin intradermal test, the typhoidin reaction, the mallein reaction and the reactions studied by Fleischner, Meyer and Shaw with Bacillus abortus and Bacillus melitensis in guinea-pigs.

It seemed that our first task, then, was to determine whether either one of these varieties of skin reactions, or perhaps both of them, could be correlated with true anaphylaxis.

Relationship of the "Immediate" Skin Reaction to Anaphylaxis. The immediate skin reaction has been used for some years as an index of safety before the injection of therapeutic horse serum in the specific treatment of pneumonia and other diseases. Its application in this connection was based primarily upon the publications of Moss; of Knox, Moss and Brown; of Longcope and Rackemann; MacKenzie and others. It should be noted, however, that the work of Moss and his collaborators, on which the assumption that the skin reaction was a reliable index of general hypersensitiveness is based, was done with rabbits, and in rabbits the skin manifestations of anaphylaxis, both intracutaneous and subcutaneous (Arthus phenomenon) are always in the form of delayed reactions, for reasons that we believe depend upon peculiarities of the rabbit rather than upon fundamental differences in the nature of the reaction. This peculiarity in rabbits, however, is being investigated in our laboratory at the present time in a study of the Arthus phenomenon, and will be reported upon subsequently. Meanwhile, it seemed to us necessary, since we were working with guinea-pigs, to determine whether in these animals an immediate skin reaction to a protein antigen, such as horse serum, egg albumen, etc., could be elicited at all, and if so whether its development was parallel to that of general protein hypersensitiveness.

In consequence, with the assistance of Dr. Woo and Mrs. Parker, who was conducting skin reactions in guinea-pigs for another purpose at the time, we carried out a number of experiments in this direction.

Series of guinea-pigs were given from 1 to 2 cc of horse serum intraperitoneally. At the end of two or three weeks they were given intracutaneous injections of horse serum in dilutions ranging from 1 in 5 to 1 in 20. Normal controls were always injected at the same time and intravenous injections of varying quantities of horse serum were made on pigs similarly sensitized in order to determine general anaphylaxis. We omit protocols, since the experi-

ments were extremely simple, and we do not wish needlessly to prolong this paper.

Our results may be stated briefly as follows: Guinea-pigs in the sensitized condition, that is, two or three weeks after the intraperitoneal injection of 1 cc of horse serum, will generally, though not always, show an immediate reaction to the intracutaneous injection of 0.1 cc of a 1 to 10 or 1 to 20 dilution of horse serum.

There is a considerable increase in the size of the wheal formed at the point of injection, the increase beginning usually within five to ten minutes and increasing up to from one-half to two hours, then gradually fading. They were usually completely gone in from two to four hours and in most cases were accompanied by a red areola quite comparable to the reaction observed in human beings under similar conditions. These reactions never caused any serious or lasting injury of the tissues.

This reaction was roughly parallel to the sensitiveness of the pigs to intravenous horse-serum injections.

It can be produced in pigs by passive sensitization, appearing on the second day after the injection of the antihorse rabbit serum almost as well as in the actively sensitized pigs.

Pigs that have been "shocked" and become anti-anaphylactic are desensitized to skin reactions made on the following day.

II. Relationship of the Delayed "Tuberculin" Type of Skin Reaction to Anaphylaxis. Having thus determined that immediate skin reactions to horse serum in guinea-pigs might be regarded, with reasonable certainty, as one of the manifestations of general hypersensitiveness, we next proceeded to investigate the same point in connection with the second type of reaction, namely, that represented by tuberculin, mallein reactions, etc. We selected tuberculosis and the tuberculin reaction as the first type to be studied, because with tubercle bacilli it is relatively easy to produce at will infections of varying intensities and different speeds of progression, and since the volume of clinical data and the careful studies of tuberculin skin reactions in guinea-pigs by Baldwin, Krause and others had cleared away a great deal of the underbrush and permitted a free approach to the basic problems. The reaction, moreover, is essentially the same type of occurrence as that following

the injection of typhoidin, mallein or abortin preparations in animals respectively infected with the organisms from which the antigens had been produced.

It is quite well known that in interpreting reactions such as the tuberculin reaction the field is fairly divided between those that hold phenomena to be manifestations of true anaphylaxis and those who deny this.

Calmette who in his recent book separates the tuberculin reactions distinctly from anaphylaxis, nevertheless seems to adhere to the general ideas of Wassermann and his collaborators in stating that tuberculous animals or man develop a "lytic" principle, presumably in the nature of a bacteriolysin which goes into reaction with the specific antigen in the tuberculin preparations, as a consequence of which poisonous substances are formed. This fundamentally, after all, is, in a sense, an "anaphylactic" conception. Friedberger has frankly regarded the phenomenon as an anaphylactic one, developing his ideas along his well-known theories of anaphylatoxin formation, basically a view identical with that of Calmette.

Serious study of the reactions from a theoretical point of view was begun by Roemer in 1909 and by Baldwin in 1910. Baldwin's work is fundamental in showing that, in spite of previous assertions, guinea-pigs could not be rendered skin-sensitive by implantation of porous filter capsules or celloidin capsules containing tuberculoprotein or living tubercle bacilli. He showed that skin sensitiveness could never be produced without actual infection with living organisms. Animals treated with tuberculoprotein, however, often showed reactions to intravenous inoculation of the homologous preparation which could be recognized as anaphylactic. His conclusions can be summarized as follows:

Tuberculous animals become sensitive to an anaphylactic test, but not uniformly so. There is no absolute relationship between the degree of sensitiveness and the stage of the disease. Injections of the tuberculoprotein may sensitize normal guinea-pigs. Sensitized guinea-pigs, however, do not react to the ordinary tuberculin test, though some respond slightly to the intradermal test. He adds, "This difference between anaphylactic sensitization and

tuberculin reactivity need not be fundamental, as it is possibly due to another factor as yet undetermined." His experiments on the transfer of passive anaphylaxis to tuberculoprotein were inconclusive, but it has been shown since then by Austrian and others that passive sensitization can be attained. From a theoretical point of view the most important observation of Baldwin is the fact that there seemed to be a discrepancy between skin sensitiveness and general anaphylaxis.

Krause, following out the work of Baldwin, confirmed and extended the above observations and established an interesting and important relationship between skin sensitiveness and the progress of infection. He asserts that skin sensitiveness develops simultaneously with the development of the initial focus, increases progressively with the lesions, varies directly with the extent and intensity of the infection and diminishes with healing. It is blunted by a general tuberculin reaction which suggests analogy to saturation, such as that which occurs in connection with anaphylaxis.

In dealing theoretically with the tuberculin reaction, especially as regards the possibility of its being an anaphylactic phenomenon, it is of great importance to determine whether or not the reaction can be passively transmitted. The evidence on this is contradictory. Only a few writers have reported positively in this connection. In 1909 Bail injected finely divided tissue mash of tuberculous organs of guinea-pigs into normal pigs, and twenty-four hours later gave the animals so treated 0.5 cc of Old Tuberculin or a preparation which in this quantity had practically no effect upon normal animals. The animals prepared with the tuberculous tissue died in some cases, while controls treated with normal tissue suspensions showed no symptoms.

Helmholz in the same year reported positive skin reactions in normal guinea-pigs two to six days after he had injected them intraperitoneally with the defibrinated blood of tuberculous guinea-pigs. Both of these observations would be of fundamental importance if they could be confirmed.

In considering the mechanism of the tuberculin reaction it will be well to examine also the work that has been done on the typhoidin reaction. Gay and Claypole believed that positive skin reactions

in rabbits were parallel with the degree of immunity of the rabbit. They succeeded in transferring the susceptibility to typhoidin from an immune to a normal animal by inoculation of 2 cc of typhoid immune serum, testing twenty-four hours later. These experiments were repeated and confirmed by Meyer and Christiansen, and in their first work with rabbits these last observers, using what they called a typhoid autolysate (by which they mean an alcohol precipitate of a heated distilled water suspension of a forty-eight-hour agar culture) concluded that "The typhoidin and similar reactions in rabbits are anaphylactic in nature and the result of an interaction of antigen and antibody." They stated that "The logical assumption from these facts is that cutaneous hypersensitiveness is the result of bacterial protein sensitization." Later, Meyer found that injected rabbits react with typhoidin more intensively than do immunized rabbits and drew the conclusion that cutaneous hypersensitiveness does not indicate that the rabbit is particularly immune and that no definite relationship existed between agglutinins and complement-fixing antibodies and skin sensitiveness. From these first two papers of Meyer we gather that he believed that in rabbits skin sensitiveness to typhoidin is a sign of infection rather than of immunity, but that, as stated in his own words, "Cutaneous hypersensitiveness of rabbits is in all probability the result of sensitization with typho or similar bacterial proteins." Nichols also considered the typhoidin reaction as he observed it on human beings as a protein sensitization.

The apparent discrepancy between the results of Baldwin with the tuberculin reaction and those of the workers just mentioned with the analogous typhoidin reaction probably depends upon the fact that Baldwin used guinea-pigs and the other observers used rabbits. When, subsequently, Fleischner, Meyer and Shaw studied cutaneous hypersensitiveness in guinea-pigs treated with repeated intraperitoneal injections of Bacillus abortus, Bacillus typhosus and Old Tuberculin, and carried out parallel experiments with animals infected with living organisms, the conclusions they reached coincide with those of Baldwin in the case of the tuberculin reaction. They found, in other words, that guinea-pigs treated with dead bacterial proteins might become anaphylactic, but did not give skin reactions.

It will be seen from this brief review of the literature that in spite of a great deal of careful work by experienced observers there is still a considerable degree of confusion. We considered it best, therefore, in studying the tuberculin reaction to begin at the bottom and reëxamine the conditions in all their details, substituting reactions with the isolated guinea-pig uterus for the intravenous test for anaphylaxis. This point is particularly important, since the intravenous injection of bacterial extracts into guinea-pigs is liable to give rise to confusing symptoms, and in carrying out such experiments we have again and again felt uncertain as to whether or not in an individual mild reaction should be interpreted as a feeble manifestation of anaphylaxis as due to toxicity of the bacteria preparation or perhaps even as the result of the injection of finely divided colloidal particles.

The first stages of our work, then, concern themselves with a study of tuberculous guinea-pigs and guinea-pigs treated with various tubercle bacillus extracts, determining in both cases the relationship of intracutaneous "tuberculin" reactions with anaphylaxis as manifested by tests with the isolated uterus. We summarize the final procedures in order to avoid a needless account of much preliminary groping for a suitable technic.

Technical Notes. The tubercle bacillus culture used throughout was a human type of moderate virulence, originally isolated, we believe, at Saranac. Many different methods of extraction were used, but the preparation with which most of the work was done was made as follows:

About 100 mg. of ground and powdered tubercle bacilli were shaken up in a shaking machine for three or four hours with 200 cc of salt solution to which had been added enough normal sodium hydroxide to give a final concentration of 0.2 per cent of the alkali. When the powder was added to this the "buffers" reduced the alkalinity which finally ranged somewhere about pH 8. After shaking, the bottles were preserved in the ice-chest and sometimes reshaken on subsequent days. The gross particles were removed before this was used, either by filtration through a Berkefeld or by high-speed centrifugation, and the slightly opalescent fluid was the basic preparation finally employed in most of the experiments and frac-

tionated subsequently for special purposes. In sensitizing guinea-pigs it was very often the unfiltered extract which was intraperitoneally injected without centrifugation.

The following method was found to be the most suitable for the sensitization of normal guinea-pigs with tubercle bacillus extracts. Young female guinea-pigs of about 150 to 200 grams' weight received from 1 to 2 cc intraperitoneally, at first on alternate but in later experiments on successive days, until ten to twelve injections had been given. It was found best to test these pigs not earlier than twenty days and usually not later than twenty-eight days after the last injection.

Skin reactions were invariably done intracutaneously with a 26-gauge needle on a tuberculin syringe, and it was attempted to inject about $\frac{1}{20}$ cc, but it was found that the concentration rather than the actual amount was a thing that seemed to count most. We gauged our skin reactions largely on the size of the wheal produced, attempting to produce the same sized wheal in comparative tests. It is next to impossible in guinea-pigs to inject absolutely accurate quantities in separate tests, but, as a matter of fact, the variations in quantity injected could never have been very great nor significant.

The uterine tests were all done in the same way, in a bath of 200 cc Ringer's solution with 0.5 to 1 per cent glucose and oxygen bubbling through the bath.

Studies with Tuberculous Guinea-pigs. The question to be answered was the following: In the course of a tuberculous infection does true anaphylaxis to tubercle bacillus products develop? and if so is it parallel in time and in intensity with the intradermal tuberculin reaction?

Guinea-pigs were intraperitoneally injected with relatively large doses of tubercle bacilli without any attempt to equalize the doses or to grade them, since we wanted to perform the test on pigs with varying degrees of tuberculous involvement and to check up by autopsy after the results had been obtained.

Skin reactions were done on these animals with the undiluted but filtered extracts described above, and with old tuberculin in dilutions usually of 1 to 5 or 1 to 10 at varying intervals from the first day after infection. When definite skin reactions appeared some of

the pigs were killed and the uteri put into the Dale apparatus, the extract mentioned above being used in most of these experiments, since in working with Old Tuberculin the margin between the non-specific action on the normal uterus and the specific action on the sensitized uterus is a relatively narrow one, a thing also found by Richard Weil, who is, as far as we know, the only one who has applied this method to tuberculosis in a report on some isolated experiments carried out in 1917.[1] A great many animals were tested in this way and the results were perfectly consistent, so that it is unnecessary to go into a detailed study of the protocols. Whenever uterine tests were done the tubercle bacillus extract used was titrated on the same day against normal uteri to make sure that we did not approach the non-specific limit, which varied considerably in these extracts. Also no skin reactions were ever carried out without being accompanied by tests, with the same material in the same quantities, on normal guinea-pigs, since in working with alcohol precipitates and other chemically manipulated substances, mild non-specific reaction will occasionally appear.

The accompanying records, Figs. 1, 2 and 3, illustrate the results obtained. Positive skin reactions were usually obtained within nine or ten days after inoculation, the earliest observed by us being between the sixth and the seventh days. At this time the animals were never anaphylactic. Graph 1 shows complete lack of anaphylactic reaction in the uterus in an animal nine days after inoculation, the day after a positive skin test had been obtained. Fig. 2 shows two uteri simultaneously put into the bath, both of them nine days after inoculation with tubercle bacilli, one of them giving an entirely negative skin reaction, the lower one giving a very marked ++++ reaction. Neither of them showed a sensitive uterus.

The third graph shows a different state of affairs, namely, an animal inoculated subcutaneously with tubercle bacilli and tested three weeks after inoculation. This animal showed a powerful

[1] Richard Weil (Jour. Am. Med. Assn., 1917, lviii, 972) obtained uterine reactions with Old Tuberculin nine weeks after injection with 0.001 cc of Old Tuberculin while the normal uterus did not react to less than 0.3 cc. The capacity of the bath is not given. He states that the reaction is an irregular one and "occurs at some stage of the disease."

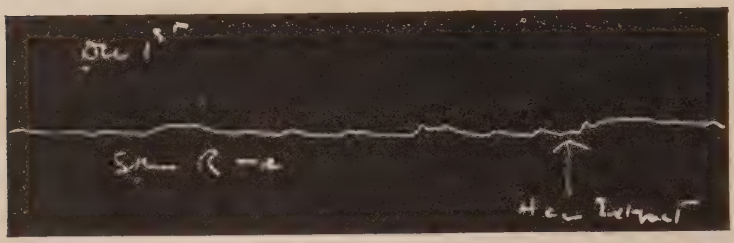

FIG. 1.—Guinea-pig 22. Another instance of positive skin reaction after nine days, with negative uterine test. This animal was of the same lot as guinea-pig 23.

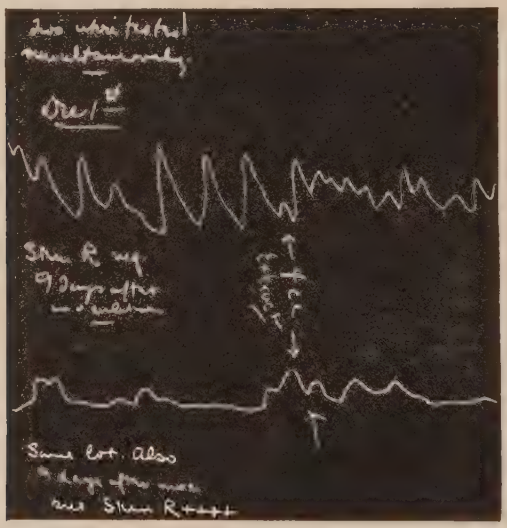

FIG. 2.—This record is a simultaneous record of two guinea-pigs inoculated on November 21, 1920, with large amounts (2 cc) of a suspension of a young tubercle bacillus culture (human). No. 23 (upper curve) inoculated intraperitoneally, and No. 24 (lower curve) subcutaneously. On December 1, 1920, skin reactions were done with two types of extract, one a sodium carbonate extract and the other a sodium hydroxide extract. No. 23, which was apparently weaker and emaciated, showed practically no reaction in either place, whereas No. 24, to our surprise, showed a definite reaction at both points. In spite of the differences in skin reactions, it will be seen that the uterine reactions in both guinea-pigs were negative. The two uteri were put into the same bath simultaneously in order to subject them to exactly the same conditions. The negative reaction of the uterus was confirmed with the other horns of both guinea-pigs. The surprising thing is the earliness with which the skin reaction appeared, which tends to confirm one of a series of observations that the skin reaction appears much earlier than the uterine reaction.

anaphylactic as well as a skin reaction. These records and a considerable number of similar ones indicate:

1. That guinea-pigs suffering from infection with tubercle bacilli may become both skin-reactive and anaphylactic.

2. That the skin reaction develops early and may exist without any detectable signs of general anaphylaxis, as evidenced by the uterine reaction.

3. That within three weeks or later, after the animals are still in fairly good condition, both the skin reaction and the uterine reaction may coexist.

It is during this period that a simulated parallelism may have led others, working with a less delicate anaphylactic method, into

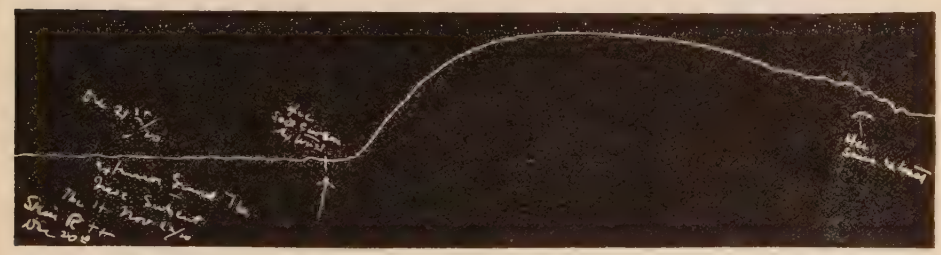

FIG. 3.—Guinea-pig 25. Inoculated subcutaneously with tubercle bacilli on November 22, 1920. Showed that powerful anaphylactic reaction may develop within three weeks after a heavy inoculation with tubercle bacilli. This, however, is unusually early. Skin reaction in this guinea-pig was positive.

confusion. Toward the end, when the guinea-pigs are moribund, both the skin reaction and general anaphylaxis may fade, another feature which simulates parallelism; but many reactions like those above have convinced us there is no question about the correctness of Baldwin's original contention that the two conditions, skin reactiveness (and probably, therefore, the other forms of tuberculin hypersensitiveness) and anaphylaxis do not necessarily coincide.

STUDIES ON GUINEA-PIGS SENSITIZED WITH MATERIALS OF KILLED BACTERIA. The next question to be answered was that concerning the conditions existing when guinea-pigs were not infected but when they were treated with products of dead tubercle bacilli. Young female guinea-pigs were prepared by the method of sensitization outlined above. They were run through in sets of six or more.

Between July, 1920, and January, 1921, five sets of such pigs were run through; all of them were treated with ten or more injections of extract. Skin reactions were done on these animals, both during the process of sensitization, that is, in the course of the ten to twenty days during which they were being treated, and at varying

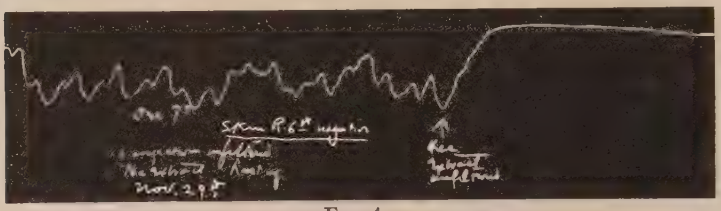

FIG. 4*a*

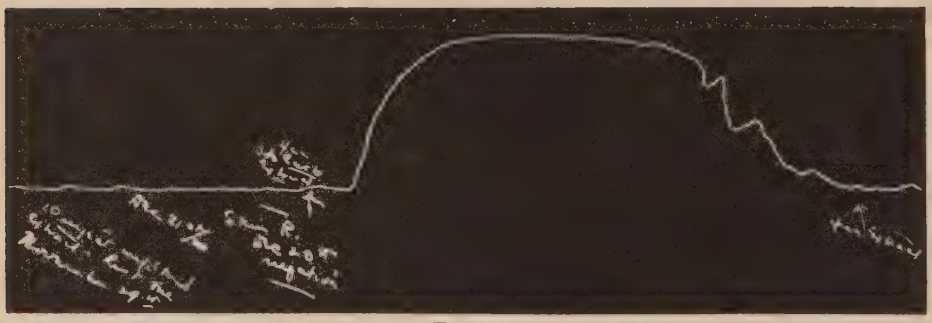

FIG. 4*b*

FIG. 4, *a* and *b*. (*a*) Guinea-pig 26. (*b*) Guinea-pig 27.—Two guinea-pigs of a series injected every other day intraperitoneally with unfiltered alkaline extract of powdered tubercle bacilli. Last injection given November 29, 1920. No. 26 gave a negative skin reaction on December 6, but a positive uterine reaction on December 7. A similar uterine test with another guinea-pig (No. 28) showed a very moderate uterine reaction, and this, with other experiments, indicates that the uterine or anaphylactic reaction does not develop until at least eight or nine days after the last preparatory injection. Guinea-pig 27 in the second record was treated exactly like the preceding animal, but here the tests were not done until December 21, slightly over three weeks since the last extract injection. Here again the skin reaction was entirely negative and the uterine test very much more powerful. The uterine reaction does not develop to its fullest extent until about two or three weeks after the last injection.

times thereafter, up to the period of three weeks after the last injection, when they were tested for uterine sensitiveness.

Practically none of these pigs showed typical delayed skin reactions comparable to the true tuberculin reaction. In only two cases did we see reactions which might have been regarded as moderate

tuberculin reactions. It may be noted that Baldwin also observed a few exceptions to his other negative results. An example of one set in which these exceptions occurred may be worth recording, since it will typify our general procedure.

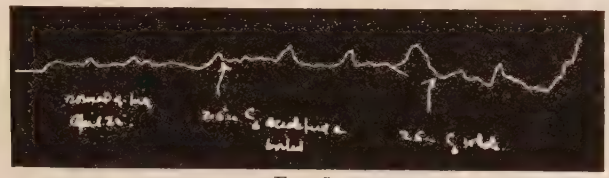

FIG. 5a

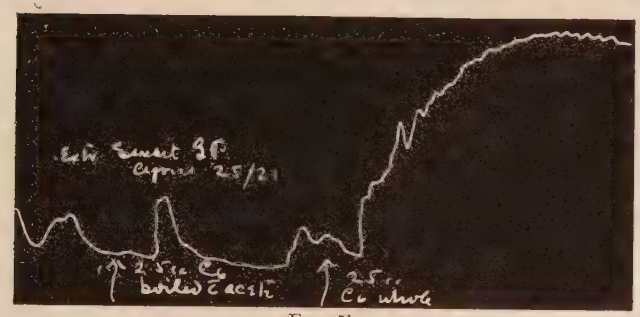

FIG. 5b

FIG. 5c

FIG. 5, *a* to *c*.—(*a*) Normal guinea-pig. (*b*) Guinea-pig 29. (*c*) Guinea-pig 30. The records show a failure of reaction of the uteri of two different sensitized guinea-pigs (Nos. 29 and 30) after the addition of the proteose residues alone, but powerful reactions when, following this, whole extracts were introduced into the bath. The record at the top is a normal control with the same extracts, showing that the reactions below are specific.

EXPERIMENTS ON THE SENSITIZATION OF GUINEA-PIGS WITH TUBERCULOPROTEIN. Six guinea-pigs, No. 323 to No. 328, were

injected intraperitoneally with 1 cc unfiltered tubercle extract as follows: September 1, 3, 7, 9, 11, 13, 20, 28. The first skin reactions were done on four of the pigs with two preparations of Old Tuberculin diluted 1 to 5 and undiluted extract similar to that injected on September 28. Negative results.

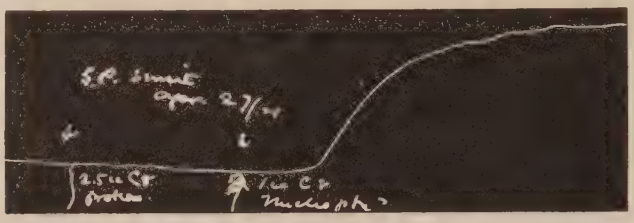

FIG. 6*a*.

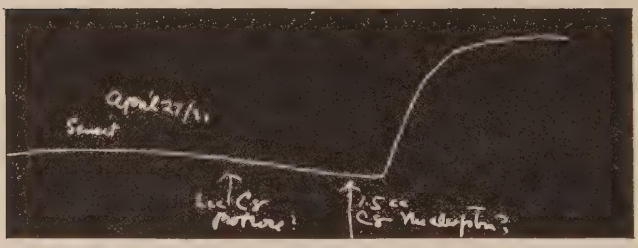

FIG. 6*b*.

FIG. 6, *a* and *b*.—This is one of two preparations in which the materials used consisted of two fractions obtained from the same 50 cc of a 0.02 per cent alkaline extract of powdered tubercle bacilli. The extract was centrifuged until clear, the supernatant fluid acidified to pH 4.5, which was the point at which the flocculent precipitate did not increase. This was centrifuged away and spoken of as nucleoprotein. This nucleoprotein was twice redissolved in more than 50 cc of alkaline salt solution and reprecipitated with acid, the material being centrifuged in each case in order to purify as much as possible. The supernatant fluid was then boiled in the acid condition to remove coagulable proteins and twice filtered through a Berkefeld filter. It will be noted that in looking at these two preparations together after testing against different strips of the same sensitized guinea-pig uterus (No. 31), the total amounts of the two fractions gave the same results, though in different proportions, an additional proof that cumulative effects had nothing to do with the results.

On October 2 all the pigs were reinjected with 2 cc of the extract and skin reactions on this day were negative.

October 7 skin reactions done on three of the pigs at the same time with four normal controls and one tuberculous control. When these reactions were read on October 8, guinea-pig No. 324 showed

that ordinarily we would have spoken of as a +++ reaction. It should be mentioned that this pig was very much emaciated and we were suspicious of its having been spontaneously infected with tuberculosis. It was killed and autopsied and a few small yellowish spots on the surface of the liver, about 2 mm. in size at the base, were taken for section and examined for us by Dr. Frederic Parker, but could not be identified as tuberculosis (we may mention that this occasional appearance of small knob-like lesions on the liver of extract-treated pigs has not been uncommon). The uterus of this pig was found to be moderately sensitive. Subsequent tests on the rest of these pigs showed only one other suspicious skin reaction, less marked than the one on No. 324.

The isolated uteri of these extract-treated pigs were with very few exceptions found to be sensitive to extract anywhere from twenty to thirty days after the last injection. Record 4 shows two guinea-pigs in which positive typical reactions were obtained with the uteri with 4 cc of a rather weak extract which in these quantities had been consistently negative.

It may be noted in passing, however, that although these extract-treated guinea-pigs did not show the typical tuberculin reaction with well-defined swollen areolæ, etc., after twenty-four hours, they did occasionally show an immediate reaction not incomparable to the immediate reaction described above for horse serum. Whenever such reactions were observed they occurred at about the time when the pigs showed anaphylactic reaction to the uterine test. This is mentioned in passing as showing that even with the products of the tubercle bacillus the two kinds of skin reaction can be elicited.

Moreover, the two graphic records shown above are only two of very many in which typical uterine reactions were elicited in pigs similarly prepared with considerably smaller quantities of various kinds of tubercle bacillus preparations. It will be seen in the lower record that a repetition of the extract showed the uterus to be desensitized, a matter which has been noted many times.

Therefore, from a considerable number of experiments similar to the ones described in perfect accord with the results obtained by Baldwin by other methods we may conclude that:

I. Delayed reactions of the "tuberculin" reaction type may

develop independently of generalized anaphylaxis in the ordinary sense in which this word is used; it may be present in tuberculous pigs before anaphylaxis to tuberculoprotein has developed, and are, with very few exceptions, entirely lacking in pigs rendered anaphylactic by sensitization with tubercle bacillus products.

These facts, then, definitely confirm the opinion, first clearly advanced by Baldwin, that "tuberculin" hypersensitiveness may develop quite independently of general tuberculoprotein anaphylaxis and that the former type of hypersensitiveness is associated particularly, perhaps solely, with the existence of an infection. This, too, is in complete keeping with the experience of Fleischner, Meyer and Shaw, who found that intradermal tests were positive only in guinea-pigs infected with the bacillus of bovine abortion, but consistently negative in those animals intensively immunized by intraperitoneal injections of dead organisms or with extracts of organisms.

II. The fundamental facts may then be stated as follows: Under the influence of contact with living tubercle bacilli, and probably other bacteria, two distinct varieties of hypersensitiveness develop in guinea-pigs. One of them, true anaphylactic hypersensitiveness, develops late and can be readily induced by appropriate treatment with dead bacterial materials, extracts, etc. The other, the typical tuberculin reaction (and probably the mallein, typhoidin, abortin reactions) may be regarded as "infection" phenomena, for while we may eventually succeed in inducing these reactions by modified methods of sensitization,[1] they are most easily and characteristically elicited as soon as an infection is established, while to date they have but rarely and atypically followed upon any form of artificial sensitization. And yet in spite of the apparent mutual independence of these two forms of hypersensitiveness, both reactions, or either one of them alone, may be elicited with one and the same bacterial extract in appropriately prepared animals.

This forces us to inquire whether a single constituent of such bacterial extracts is responsible for both types of reactions or whether it is possible to separate from such preparations two functionally distinct fractions, one capable of eliciting the typical

[1] See below.

anaphylactic response and the other active in the "tuberculin" sense.

The latter opinion is rendered likely by the generally accepted view that the true anaphylactinogens are proteins, whereas most of the work done with "tuberculin" seems to indicate that these substances are of a simpler structure.

Loewenstein and Pick,[1] among others, conclude that the active substance in tuberculin is a protein-free, alcohol precipitable substance which is not coagulated by boiling either in neutral or acid reaction, cannot be precipitated with ammonium sulphate, gives a negative Biuret reaction, but is precipitated with tannic acid. We will not go into the details of their analysis, but their final conclusion is that the active substance is not a protein but belongs in the class of protein-split products which Fischer has spoken of as "polypeptides." They claim that dialysis of the concentrated tuberculin solution weakens it after a short time, and that after prolonged dialysis its activity is entirely lost; they do not, however, state the nature of the membranes used nor the particular conditions under which dialysis was performed. The active tuberculin substance, moreover, which Calmette uses for his ophthalmo reaction is the white, flocculent and highly soluble substance which is precipitated out of tuberculin solutions upon the addition of ten to twenty volumes of absolute alcohol.

Along the lines of these investigations we proceeded to attempt separation of the various substances which might be obtained from tubercle bacillus extracts.

Attempts at Chemical Separation of the "Tuberculin" from the Anaphylactinogen. From powdered tubercle bacilli of the human type kindly furnished to us by Dr. Petroff, of Saranac, we produced extracts by shaking in a 0.02 per cent NaOH solution in physiologic salt solution. Three or four hours' shaking and perhaps a day or so in the ice-chest sufficed to bring a considerable amount of the material into solution. This extract was centrifugalized until all the particles had been removed and a moderately opalescent supernatant fluid was decanted.

This material, upon being acidified to approximately pH 5 to 6,

[1] Biochem. Ztschr., 1911, xxxi.

with 2 per cent acetic acid in the cold, became turbid and soon precipitated in large flakes. Further acidification up to pH 4 did not redissolve these flakes. This precipitate represented the bulk of the dissolved substances in the extracts. The precipitates could be redissolved in a slightly alkaline salt solution and reprecipitated with acid. Because of their precipitability by acetic acid in the cold we designated these substances as "nucleoproteins" or phosphoproteins, although we do not wish to commit ourselves chemically, since we are quite aware of the indefiniteness of these biochemical terms and realize fully our incompetence to deal authoritatively with a chemical problem of such difficulty without further intensive study.

After removal of these acid precipitable substances by centrifugation and filtration through Berkefeld candles the fluid was brought to a boil in the acid condition, and sometimes a very faint turbidity developed which was taken to represent coagulable protein, albumin or globulin, or both. These precipitates were so fine and slight that only Berkefeld filtration would remove them, and even this was not always completely successful.

The fluid was then neutralized, that is, brought to an approximate pH of 7.

When this neutral fluid was filtered hot we observed on numerous occasions that a slight precipitate developed overnight in the ice-chest and that this precipitate redissolved on heating, a point which indicated the possibility that the tubercle bacilli may contain Bence-Jones protein. This point, however, will need further chemical analysis, a task which we have not yet had time to undertake. The water-clear fluid which was left after removal of all these substances gave in all cases a very definite precipitate with alcohol. This precipitate could be thrown down, collected and redissolved in water with salt solution, and like the similar precipitate obtained from crude tuberculin was astonishingly soluble.

This final water-clear material gave no Biuret reaction, usually gave no sulphosalycilic reaction, though occasionally this was very faint; in most cases it gave no Millon reaction, was not clouded on boiling with acid and usually gave no xanthroproteic reaction, though in some cases a slightly yellowish color appeared when the

ammonia was added in the second part of the test. This material, for want of a better name, we speak of as our "proteose" residue.

Having thus fractionated the original tubercle bacillus extract we proceeded to carry out biologic reactions with various parts, comparing chiefly the whole extracts with the dissolved substances which precipitated with acid in the cold (nucleoproteins) and with the final solutions from which the acid precipitable, acid and heat coagulable substances and the material suspected of being Bence-Jones protein had been removed (proteose residue).

We have done a large number of skin reactions on tuberculous guinea-pigs with the apparently protein-free "proteose" residue, always controlling with tests on normal guinea-pigs. This precaution is absolutely necessary, since we have found (as we were warned to expect by Baldwin) that repeated alcohol precipitation or prolonged boiling in an excessively acid reaction (pH 4 or below) may eventually lead to non-specific toxicity both for the skin test and for the isolated uterus.

Invariably this final "proteose" material gave specific skin reactions, often quite as powerful and never more than slightly less than the original whole material. And, indeed, it was to be expected that a certain amount of the tuberculin active substance might be carried down in the flocculent precipitate formed when the acid is first added, thus depriving the solution of a certain percentage of its activity. The following protocol is an example of experiments of this kind.

SKIN REACTIONS READ AFTER TWENTY–FOUR HOURS

Tuberculous guinea-pigs.	Whole extract, 1 to 2.	Proteose, 1 to 2.	Whole extract, 1 to 5.	Proteose, 1 to 5.
No. 1125	++++	± ± ± ±	± ± ±	± ±
No. 1149	+++	++	+++	+++
No. 1102	+++	++	+++	++

	Whole extract undiluted.	Proteose undiluted.
No. 1174	++++	+++
No. 138	+++	+++

Two normal controls show no non-specific reactions.

Reactions of this nature, often repeated, indicate definitely that in removing the acid precipitable and the coagulable protein substances from the tubercle bacillus extracts we did not remove any considerable part of the skin reactive substance. Moreover, they justify the assumption that in the "tuberculin" skin reactions the antigen need not be a protein substance but probably belongs in the category of the so-called "proteoses" or perhaps "polypeptides."

It must be stated, however, that the "nucleoprotein" fraction precipitated from the original extracts with acid in the cold always retained "tuberculin" activity. In spite of repeated reprecipitation and resolution we have never succeeded in entirely removing the capacity of inducing skin reactions of the "tuberculin" type from these acid precipitable substances. This may be due to the absorption of the "proteose" material by the heavy flakes of the precipitate. On the other hand subsequent experiments suggested the possibility that these "nucleoproteins," which constitute the bulk of the soluble material of the bacterial cell, might represent the mother substance from which the "proteose" materials are derived.

However this may be the proteoses alone are fully capable of giving rise to the typical tuberculin reaction in the infected guinea-pigs.

This being the case, what is the effect of this "proteose" fraction upon the isolated uterus of the "extract" sensitized or anaphylactic guinea-pig in which no tuberculin skin reaction can be elicited?

In these experiments our results were often not as clean cut as they might have been. Nevertheless, in a considerable number of tests there was indication that the "proteose" fraction, powerfully skin reactive, had little or no effect upon the uteri of extract-sensitized animals, which contracted powerfully when brought in contact with the whole extracts or the acid-precipitable fractions in amounts that had no effect upon the normal uterus.

Two such records are attached.

Record V shows the uterus of a pig, sensitized with extract, which was unaffected by 2 and 2.5 cc of the proteose fraction, but contracted powerfully and went into spasm when 1.5 cc of the whole extract was added to the bath.

Record VI shows a similar experiment in which 2.5 cc of the

proteose fraction had no effect, whereas as little as 1 cc of the acid precipitable fraction produced a strong contraction and spasm.

Six as sharp and unambiguous as the above were obtained. But in addition we must state that on three occasions the proteose material contracted the uteri of highly sensitized and of tuberculous guinea-pigs when relatively large amounts of "proteose" were used. Whether this means that we had not freed the "proteose" completely of protein or whether it signifies that, with sufficiently energetic treatment, the proteoses, too, may form antibodies, is a question that cannot be definitely answered. We incline to the latter view for reasons that will be discussed below.

Notes on the Dialysis of Tuberculin. It will be remembered that the chief clinical difference between the "immediate" skin reaction of the "anaphylactic" type and the tuberculin type of reaction is the fact that the former consists in a rapidly developing urticaria-like swelling which disappears in a relatively short time and leaves little or no injury behind. The latter, however, comes on slowly, and when once developed is accompanied by manifestations of inflammation and eventually profound injury of cells, often with cell death, a state of affairs which at least suggests the penetration of the injurious substances into the cells themselves. This, taken together with our knowledge of the relatively simpler structure of the "tuberculin" substances and Loewenstein and Pick's claims concerning their diffusibility, suggest a number of interesting lines of reasoning.

Is it not possible that the diffusibility of antigenic substances is a very important factor not only in the reaction of these diffusible substances with the cells but also in the formation of antibodies? Is it not likely that substances which are practically non-diffusible should be excluded from direct reaction with the tissue cell and that the mechanism of antibody formation is a device for reaction with non-diffusible materials? And if this is so, is it unreasonable to suppose that the reaction of the body to substances which are relatively more diffusible should become increasingly intracellular as diffusibility increases, and that antibody formation, in the usual sense of the word, should therefore become less and less essential as relative diffusibility increases? Such a view would explain at the

same time the apparent intracellular nature of such reactions as the tuberculin reaction and the difficulties encountered in attempts at passive transfer of such sensitiveness. Accordingly, we thought it worth while to attempt to separate by dialysis the substance which produced the tuberculin skin reaction from that which caused typical anaphylaxis.

In planning experiments upon the diffusion of antigenic substances through semipermeable membranes we are quite aware of the many pitfalls which entrance into such a field on the part of a biologist implies. We know that it is by no means proved that the living cell membrane is at all permeable to substances like the proteoses and that experiments with dialysis *in vitro* may lack some of the essential criteria that govern dialysis in the animal body. Nevertheless, we thought it worth while to investigate the diffusibility of the "tuberculin" active substances. We at first tried to work with membranes graded by the methods of Brown and of Gates. But since the differences in diffusibility of proteins and the "proteose-like" substances with which we are concerned can be very slight ones only, we abandoned this and worked empirically, attempting in many tests to find membranes which would "let through" the tuberculin active substances but "holdback" the anaphylactinogenic ones.

Our general procedure consisted in taking various tubercle bacillus preparations, the skin reactiveness of which we had determined on tuberculous guinea-pigs, subject them to dialysis either in celloidin or in fish-bladder bags, in a closed system, testing the material outside of the bag from time to time on tuberculous and normal guinea-pigs. As soon as the "outside" material gave indication of containing the skin reactive substance the same material was used upon a sensitized and a normal uterus. In a few experiments we readily obtained results which at least indicate that the tuberculin active substance is more readily diffusible than the anaphylactinogens.

Experiment I. A glycerin salt solution extract of powdered tubercle bacilli was started on February 7. One gram of powdered tubercle bacilli was infused in 500 cc of a 5 per cent glycerin salt solution, heated to 70° for one hour and placed in the incubator overnight. It was then shaken for several hours on consecutive

days. This material was then simmered down to 150 cc at a temperature of 75° to 80°, never quite being allowed to come to 80°, and was then filtered through paper. It gave a strong sulphosalicylic acid reaction.

On February 15, 40 cc of this material were placed in a fish-bladder bag in a closed system with 40 cc of salt solution outside.

On February 17, skin reactions were done with the material inside and outside the bag, and it was found that a very definite though mild skin reaction resulted from the material outside the bag. The relative strength of the skin reaction "inside" and "outside" the bag are given in the following table:

READINGS ON SKIN REACTIONS DONE FEBRUARY 17

Guinea-pigs.	B_2 outside.	B_2 inside.
No. 352	++	++++
No. 353	++	++++
No. 354	++	++++
No. 351	−	++++
No. 332	±	++
No. 336	±	++
No. 1132	+	+++
No. 1132	+	+++
Normal control	−	±
Normal control	−	±

++++ violent and large. ++ distinct, unmistakably moderate.
+ distinct but faint.

On February 18, this material was tested upon normal uteri and it was found that 2.5 cc of the material and the material outside the bag added to a 200 cc Ringer bath, in which a normal uterus was rhythmically contracting, produced no marked changes in the rhythm. The material was then tested on the uterus of a guinea-pig sensitized as above described with tubercle bacillus extract. As will be seen in the accompanying graph, 4 and 6 cc of the outside of the bag, which gave a skin reaction, produced no spasm of the sensitized uterus, whereas 2 cc from the inside of the bag gave a typical and marked reaction (Fig. 7). We realize that this may have been a quantitative result, due to a greater sensitiveness of the skin than of the uterus. This does not seem likely under the circumstances, but, nevertheless, must be considered.

We have mentioned above that Loewenstein and Pick claimed that they could completely free Old Tuberculin of its activity by prolonged dialysis against running water. We have tried this a great many times but never entirely succeeded. We did obtain one experiment in which the contents of a fish-bladder bag lost skin reactiveness and retained anaphylactic antigenic properties for the uterus. There was, however, in this case considerable bacterial growth which was hard to avoid when running water was used, and the apparent separation of the skin reactive substance from the Old Tuberculin in the bag may have been due to bacterial destruction of this substance. The activity of this material on the uterus

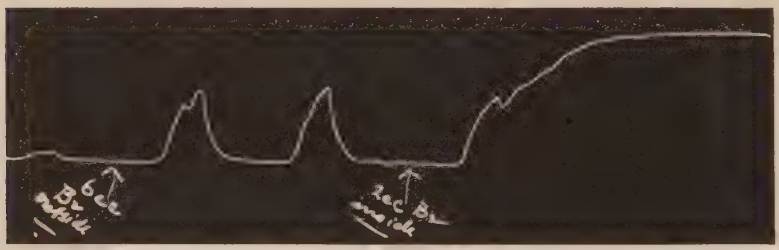

FIG. 7.—February 18. The sensitized guinea-pig treated with Material B_2 which, as stated before, gave definite though somewhat weakened skin reactions. Here, 6 cc on the outside of the bag, which still gave skin reactions, gave no anaphylactic reaction. The high rise is probably due to the fact that this uterus was very irritable. That the rise was not an anaphylactic reaction is apparent from the immediate return to the base-line, and the fact that there was no apparent desensitization to the subsequent addition of 2 cc from the inside of the bag. Dialysate was positive on skin and negative on uterus.

was not due to histamine formation, since it was inactive against the normal uterus. We believe that our failure to free the tuberculin preparations from the skin reactive substance by prolonged dialysis against running water may have been due to precipitation of substances inside the bag before dialysis was completed, but at any rate we could never entirely repeat Loewenstein and Pick's experiments.

The dialysis experiments will be continued with more elaborate methods at a later date. For the present they are mentioned only for the sake of comparison.

We have noted above that Loewenstein and Pick claimed that

they could completely free Old Tuberculin of its activity by prolonged dialysis against running water.

For this reason we determined to try the reverse experiment, that is, attempting by prolonged dialysis to remove the skin reactive substance entirely from the inside of a dialysing bag, leaving the anaphylactinogenic substance behind. We tried this with various preparations, but never succeeded even with prolonged dialysis to free the materials on the inside of the bag entirely of the skin reactive substance, although such a result had been claimed by Loewenstein and Pick. Finally, since we know from the experiments of Richard Weil and from some of our own earlier experiments that Old Tuberculin still contains some of the anaphylactinogenic substances, we placed 25 cc of concentrated New York Board of Health Old Tuberculin in a fish-bladder dialysing bag and dialysed this against running water. The Old Tuberculin we used still gave powerful sulphosalycilic acid reaction for protein and gave a definite precipitate on being acidified with acetic acid in the cold after dilution to about 1 to 20. The precipitate did not redissolve upon heating but was rather intensified. With this preparation we had what we must now consider a rather fortunate accident. Because we were dialysing with running water we could not carry out the procedure in the refrigerator and bacteria began to grow in the dialysing bag. In the course of five days the material from the inside of the bag gave practically no skin reaction on tuberculous pigs as compared with Old Tuberculin diluted 1 to 3, which corresponded to approximately the dilution which had been attained by the substance dialysed in the course of dialysis. This material, which no longer gave skin reactions, was tested against normal and sensitized uteri, and it was found that although 3 cc of the material gave no reaction on the normal uterus, powerful reactions were obtained in quantities as low as 0.5 cc and a weaker but still noticeable reaction obtained with 0,2 cc. The possibility that substances like histamine were formed by the bacteria and account for the uterine reaction can be excluded by the failure of this material to affect the normal uterus. Whatever may have been the reason for this result, whether dialysis had anything to do with it or whether it means simply that the bacterial growth had destroyed the "tuberculin" substances while

leaving the anaphylactinogens intact, it seems a definite and sharp separation of the two functions.

III. Neither chemical fractionations nor the diffusion experiments furnish absolute proof that we can completely separate the anaphylactinogen from the "tuberculin" active substances. Nevertheless, both series of experiments point in this direction, and, taken together with the mutual independence of the two types of reaction in guinea-pigs, render such a conception an extremely likely one.

However this may eventually prove to be, one fundamental fact remains definite, namely, that the "proteose" fraction alone can elicit the particular form of hypersensitiveness which we speak of as the "tuberculin" reaction, and that the potency of the proteose residue in this respect is but slightly less than that of the whole extract in spite of the fact that proteins have been removed as completely as this is possible by boiling with acid.

Assuming, then, that the "tuberculin" type of reaction is a response to a "proteose" antigen while the anaphylactic reaction is associated with the proteins, we are still confronted with the very puzzling fact that the "proteose" reaction occurs only in infected animals and cannot ordinarily be induced in animals treated and rendered "anaphylactic" by injections of dead bacterial substances.

It might be assumed that the living tubercle bacillus in contact with the animal tissues produces a sensitizing substance which is not present in culture fluids or in dead tubercle bacilli. This idea is rendered improbable by the consideration that although we almost uniformly fail in sensitizing guinea-pigs to the cutaneous test by prolonged treatment with concentrated culture fluids or with bacillus extracts, nevertheless skin reactions can be elicited in tuberculous animals by the proper application of such substances.

Another possibility, however, is that the substance which sensitizes to the tuberculin reactions is actually represented in the various tuberculin preparations and tubercle bacillus extracts, but that the intermittent method of injection which must necessarily be used in the preparation of guinea-pigs or other animals does not simulate the manner in which these substances are being constantly diffused out into the animal tissues from growing foci. The supposition that the difference may lie in the manner of sensitization, both as to the

time factor and in regard to the quantitative relations, is suggested by such experiments as those of Krause, who has shown that cutaneous hypersensitiveness coincides with the establishment of a focus, diminishes with the healing of the focus and varies directly with the intensity of the disease. It is suggested that in specific phenomena of hypersensitiveness of the tuberculin, typhoidin, mallein types, etc., we may be dealing with antigens that are subject to laws of sensitization and antibody formation quite different from those governing the phenomena of protein anaphylaxis—in which the hypersensitiveness is elicited only by an intensive and concentrated contact with the antigenic substances and in which, soon after the stimulus is removed, the hypersensitiveness diminishes. This, at least, seems a logical conclusion in regard to guinea-pigs.

It should also be borne in mind, as has been suggested by Krause, that a general tuberculin reaction blunts skin hypersensitiveness considerably and that a similar blunting of both the anaphylactic and the skin hypersensitiveness has been noted by others, as well as by ourselves, in the late stages of a fatal tuberculosis in guinea-pigs, observations which indicate a reaction at least analogous to antigen-antibody reactions in general.

These considerations suggested to us the possibility that although we might be giving the animals which we had treated with bacterial extracts some of the material which sensitizes them in the course of infection, we were not, perhaps, administering to them a sufficient amount of this proteose-like substance and were not giving it with the continuity with which it passes into the circulation of an animal suffering from an active process.

In order to obtain some light upon this we carried out a number of experiments as follows:

Tubercle bacillus cultures were grown on 5 per cent glycerin peptone broth. When the growth had reached the size of a silver dollar or slightly more about one-quarter of this growth was carefully lifted into another flask containing 100 cc of similar glycerin broth and the remaining three-quarters were killed at 80° for one-half hour. This dead growth, at least three-quarters of the original bulk of tubercle bacilli, was now washed several times with salt solution and infused in a flask containing 100 cc of broth of the same lot as

that on which the living one-quarter had been inoculated. The two flasks were then put into the incubator and after the second day skin reactions were done every day with the fluids from this broth on tuberculous guinea-pigs.

The accompanying figure shows the results (Fig. 8). The reaction marked "1" on this figure is the reaction obtained with 1.0 cc intracutaneously injected of a 1 to 3 dilution of the four days'

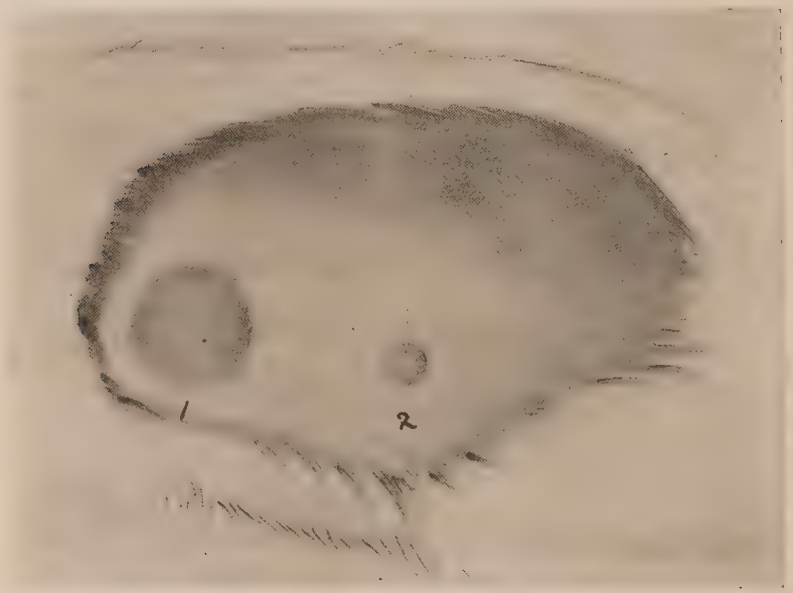

FIG. 8.—Drawing of skin reactions on a tuberculous guinea-pig, carried out with broth from a culture of living tubercle bacilli four days old; 1: 3 dilution, at the point marked 1, and a similar reaction with a similar amount of broth from a killed culture, 1: 3 dilution, at the point marked 2.

growth in a highly sensitive guinea-pig. The reaction marked "2" shows the corresponding reaction carried out with the broth in which the dead tubercle bacilli, three times greater in bulk than the living ones, had been infused for the same length of time and used in the same dilution.

In order to eliminate any obstruction to diffusion which might have been brought about in the dead culture by the fact that heat

was used in killing, we did another experiment in which ½ per cent carbolic acid was added to the culture material to kill it.

Such an experiment is the following one: In this case a living growth about 1.2 cm. in diameter was floated on a flask of 100 cc glycerin broth and the remainder, about four times this amount, was submerged by shaking in 100 cc of a similar broth flask and 0.5 per cent carbolic acid added. On the sixth day reactions were done on tuberculous guinea-pigs and on a control, with the following results:

Tuberculous guinea-pigs.	Living.		Killed.	
	1 to 10.	1 to 20.	1 to 10.	1 to 20.
No. 598	+	+	+	±
No. 1066	++	+	+	±
No. 1053	++	±	−	−
Normal control	−	−	−	−

Reactions of this type would indicate that the materials which caused skin reactions of the tuberculin type were being constantly diffused out from the growing cultures while a limited amount only could be extracted from dead tubercle bacilli.

It is not impossible also that such a conception might indicate that these proteose-like materials were constantly being produced from the more complex material which we have spoken of tentatively as "nucleoproteins," which represent the bulk of the soluble bacterial constitutents, and that this is the reason why we have never been able to free these substances of their skin-reaction capacity.

With this in mind we have recently treated four guinea-pigs with large amounts of the so-called "nucleoproteins," giving each animal four intraperitoneal injections, each of which represented at least 50 cc of original extracts. And in these animals, fourteen days after the last injection, we saw the first hopeful indications of positive skin reactions of the tuberculin type artificially induced. These experiments will be continued.

IV. The work reported in the preceding sections justifies, we think, a number of definite conclusions. In addition to this some of the experiments indicate a line of thought which may lead to considerable alteration in our conceptions, both of phenomena of bacterial hypersensitiveness and of infection:

1. In guinea-pigs two fundamentally different types of intra-

dermal reactions may be observed. One of these is the immediate transitory reaction which develops in animals sensitized against proteins (horse serum, etc.) and may be regarded as one of the manifestations of general protein hypersensitiveness or anaphylaxis; the other is the "tuberculin" type of skin reaction which develops more slowly, leads to a more profound injury of the tissues and is independent of anaphylaxis as ordinarily conceived.

2. The "tuberculin" type of hypersensitiveness (as well as probably the typhoidin, mallein, abortin reactions, etc.) does not develop at all in guinea-pigs sensitized with proteins, like horse serum, etc. While this form of hypersensitiveness may eventually be induced with materials not bacterial in origin it has been observed up to date only as a reaction of bacterial infection.

3. Methods of treatment with protein material from bacterial cultures which sensitize guinea-pigs to anaphylactic reactions with the bacterial extracts do not sensitize them to the "tuberculin" type of reaction. Such sensitization is easily accomplished only by infecting the animals with living organisms. No reliable method of sensitizing guinea-pigs to such reactions with dead bacterial material has as yet been worked out, though a few hopeful experiments have been obtained with massive injections of large amounts of the acid precipitable substances (nucleoproteins?) from bacterial extracts.

4. In animals made hypersensitive to the "tuberculin" type of reaction by infection with living bacteria the reaction may be elicited by intradermal injections of bacterial extracts from which all coagulable proteins, nucleoproteins and Bence-Jones proteins have been removed, as well as this can be done, by boiling with acid, etc. This "proteose residue" alone suffices to elicit such reactions. The exact chemical nature of this so-called "proteose residue" must be further studied and analyzed when we have had opportunity to produce bacterial extracts in large quantity.

These points seem incontrovertible on the basis of our own experiments as well as those of other workers.

There thus seem to develop two definite forms of hypersensitiveness in guinea-pigs infected with bacteria, typical anaphylaxis in which the protein material of the bacterial cells are concerned,

which develops late and which can be induced by repeated injections of dead bacterial material, and a hypersensitiveness to non-protein constitutents which differs from the former both in the laws that govern sensitization and in the manifestations which follow injections into the sensitized animals.

While there is virtual agreement among immunologists concerning the essential mechanism of protein anaphylaxis, its dependence upon an antigen-antibody reaction, and the dominating role played by the sessile antibodies, the mechanism of hypersensitiveness to tuberculin and similar bacterial substances is still a problem of much uncertainty.

The most striking difference between the two phenomena lies, as we have seen, in the criteria of sensitization, in that hypersensitiveness to the "tuberculin" type of reaction can hardly ever be induced by any of the ordinary methods of preparation with the constituents of dead bacteria but develops promptly (seven to ten days) in the course of actual infection with living organisms.

The considerable specificity of such reactions forces the conclusion that the sensitizing substance must in some way be derived from the infecting microörganisms.

The idea that the failure of sensitization with dead culture materials is perhaps due to the elaboration in the body of infected animals of bacterial products not represented in extracts of test-tube cultures is rendered unlikely by the fact that in the "tuberculin-sensitive" infected animals we can produce the reactions by the application of such dead extracts. It is neither logical nor in keeping with biologic experience to assume that one substance will sensitize to reaction with another. (This mistake was made early in the study of anaphylaxis in another connection and caused considerable delay of progress.)

Krause has shown that tuberculin sensitiveness may be blunted in infected animals by massive but sublethal injections of tuberculin, and we have obtained some indications of the same thing. Moreover, others as well as ourselves have seen tuberculin reactivity decline in guinea-pigs and in man in the stages of very severe infection. These facts would eliminate any assumption of mere

cumulative injury as explaining this type of reaction and stamp it as a mechanism at least analogous to ordinary anaphylaxis.[1]

The only remaining possibility to explain the difference between infected animals and those treated with dead bacterial constituents would be to assume that the difference must lie in the manner in which the sensitizing substance is administered to the animals and that sensitization with the "proteose residue" materials depends upon criteria of sensitization, differing in regard to the time and quantity factors from those governing protein sensitizations. If one considers the relatively simpler chemical structure and perhaps physically greater diffusibility of the materials concerned in this reaction one might readily expect such differences in the methods needed for sensitization.

In keeping with such a line of reasoning our experiments have shown that the "tuberculin" active materials are constantly and rapidly being diffused out into the culture fluid from growing organisms in quantities greater than can be extracted from similar amounts of the dead bacteria. It seems reasonable to assume from this that the same thing may happen in the animal body harboring a growing focus. And it would seem quite likely that the association of the "tuberculin" type of reaction with actual infection may depend upon the fact that sensitization to these non-protein substances depends upon a constant steady absorption of large amounts of the material.

Moreover, the only hopeful experiments on the artificial production of tuberculin sensitiveness in guinea-pigs obtained by us were those in which massive doses of the "nucleoprotein" material injected into guinea-pigs gave rise to a moderate skin sensitiveness.

Does the so-called "proteose residue" form antibodies, and if so, are substances analogous to antibodies involved in the tuberculin type of hypersensitiveness?

The failure to passively transfer this form of hypersensitiveness to normal animals with the blood and tissues of tuberculin sensitive ones would suggest that no antibodies are involved. But this is not conclusive on the basis of available experimental facts. We

[1] Direct attempts to show such cumulative toxic action have failed in our hands.

are inclined to believe that antibodies of a sort are involved for the following reasons:

1. In our experiments with the uteri of highly sensitive extract-treated guinea-pigs and of tuberculous guinea-pigs we have occasionally had positive reactions when the "proteose residue" alone was used.

2. We believe that these "proteose" substances are entirely analogous to the substances studied by Avery and Dochez in the urine and blood of typhoid and pneumonia patients. They obtained precipitin reactions against homologous immune sera with the urine of infected cases, concentrated by evaporation after boiling with acetic acid to remove coagulable proteins.

3. Petroff, with whom we discussed this "proteose residue" early in our work, has produced it and tells us that he has obtained precipitin reactions with it by titrating it against the serum of a sheep treated for a long time with tubercle bacillus products.

In suggesting an antibody response to a non-protein antigen we are aware that we are flying in the face of what has been regarded as a well-established doctrine of immunology. This is justified, or at least mitigated, we believe, by the consideration that reactions of the antigen-antibody type are the only explanation of specificity, and tuberculin, mallein and typhoidin reactions are to a considerable degree specific. If such reaction bodies cannot be produced by precisely the same methods of administration as to time and quantity which are successful in calling forth protein antibodies, this should not astonish us, since after all the substances we are dealing with are simpler in chemical structure than are the proteins and physically are probably of relatively greater diffusibility. It may be that the greater diffusibility of the "proteose-like" substances transfers much of the actual reaction phenomena to an intracellular location, and that this to some extent influences the presence of circulating antibodies. It may also be that these more diffusible non-protein antigens are more rapidly eliminated from the animal body than are the proteins. Indeed, the above-mentioned observations of Avery and Dochez, and the recent work of Wilbolz, Lanz, Imhof and Gibson and Carroll, who demonstrated tuberculin active antigens in the urine of active cases, would corroborate such a view. The

evidence available at the present time, however, concerning antibody formation to these non-protein substances is, we recognize, largely indirect, at least as far as our own work is concerned, and we present it in the present connection purely as a working hypothesis.

Finally, perhaps the most important theoretical consideration indicated by our experiments is the following: We have in the tuberculin reaction a form of hypersensitiveness which seems to be (in guinea-pigs at least) analogous entirely to the typhoidin reaction, the mallein reaction and the abortin reaction. Whenever reactions of this type have been carefully studied, whatever the bacteria involved, they have been associated with infection, as in tuberculosis, and have been followed by analogous clinical manifestations. It would seem perhaps that we are dealing with a law applicable to bacterial infection in general.

It would appear that certain non-coagulable substances of uncertain chemical constitution are being constantly elaborated in the course of bacterial growth and passed into the circulation of infected animals. As a result of this infected animals become sensitized to these heat and acid resistant materials in tuberculosis in the course of one or two weeks, in the case of more rapidly growing bacteria perhaps sooner. Early in the course of infection the animal becomes sensitized and subsequently the further elaboration and distribution of these materials from the bacterial focus play a fundamental part in the injury of the animal. These "proteose-like" substances, like tuberculin, possessing but slight toxicity for the normal animal, become highly toxic to the sensitized one. Thus these substances, while not being true exotoxins in the ordinary sense, would still represent a highly toxic bacterial product comparable in its injurious effect to toxins when produced in the body of an animal thus sensitized.

If there is any value in these deductions the attention of bacteriologists should be turned to the non-protein constituents of bacterial cells in their further immunological studies as well as to the protein materials.

It is obvious that the next step in our investigations must consist in producing the non-coagulable material from bacterial extracts in considerable quantity to determine their antibody forming

properties in detail and elucidate, if possible, the laws which govern sensitization with them. This work has been begun, but it has seemed advisable to publish this as far as we have gone because it will take a long time before it can be completed.

REFERENCES

Zinsser and Parker: Jour. Exper. Med., 1917, xxvi, 411.
Fleischner, Meyer and Shaw: Am. Jour. Dis. Child., 1919, xviii, 577.
Moss: Jour. Am. Med. Assn., 1910-1, v, 776.
Knox, Moss and Brown: Jour. Exper. Med., 1910, xii, 562.
Longcope and Rackemann: Jour. Exper. Med., 1918, xxvii, 341.
Leake and MacKenzie: Jour. Exper. Med., 1921, xxxiii, 601.
Calmette: L'Infection bacillaire et la tuberculose, Masson, Paris, 1920.
Roemer: Berl. klin. Wchnschr., 1909; Deutsch. med. Wchnschr., 1909.
Baldwin: Jour. Med. Research, 1910, xxii, 189, with Kruse: Stud. Saranac Labor., 1910.
Krause: Am. Rev. Tuberc., 1917, i, 65.
Bail: Ztschr. f. Immunität., 1910, iv, 470.
Helmholz: Ztschr. f. Immunität., 1909, iii, 371.
Gay and Claypole: Arch. Int. Med., 1914, xiv, 671.
Meyer and Christiansen: Repr. G. W. Hooper Foundation, 1916–1917, ii, 1.
Meyer: Repr. G. W. Hooper Foundation, 1916–1917, ii, 68.
Nichols: Jour. Exper. Med., 1915, xxii, 780.
Loewenstein and Pick: Biochem. Ztschr., 1911, 31, 142.
Brown: Biochem. Jour., 1915, ix, 591.
Gates: Jour. Exper. Med., 1921, xxxiii, 25.
Wilbolz: Correspond.-Blatt. f. Schweiz. Aerzte, 1919, xlix, 793.
Lanz: Schweiz. med. Wchnschr., 1920-1, 321.
Imhof: Schweiz. med. Wchnschr., 1920-1, 1033.
Gibson and Carroll: Jour. Am. Med. Assn., 1921, lxxvi, 1181.
Austrian: Johns Hopkins Hosp. Bull., 1912, xxiii, 1.
See also the excellent summaries of:
Doerr: Kolle and Wassermann's Handb., 1913, ii, 947.
Coca: Tice's Practice of Medicine, New York, Prior & Co., 1920, p. 107.
Wells, Gideon: Physiological Review, 1921, i, 44.

DISCUSSION

Dr. Simon Flexner, New York: I am sure that nobody in the audience envies my position at this critical moment, having listened to these three remarkable and comprehensive papers on the subject of the discussion. Surely very little is left to be presented to you.

I have been deeply interested in the general biologic point of view which has dominated this discussion before an audience composed of medical men. This fact in itself is indicative of the growth of the science of medicine which is going on everywhere today. As I look upon the complex processes described I view them not as isolated happenings in nature but as something

for us to affiliate with biologic phenomena in general and to ascertain the manner in which the more general biologic knowledge of the time is to be used in interpreting intricate reactions taking place between the host on the one hand and the parasite on the other.

Anyone who has undertaken to unravel some of these reactions is impressed by their complexity, but we are, I think, moving in the direction of simplifying our material so that we can handle it more effectively. There are two or three particular points to which I wish to allude: One often hears that ethnologists are getting concerned lest pretty soon they will have no new fields to explore. So few primitive peoples are left to study that unless they make haste they will no longer be able to procure material dealing with primitive culture. Similar conditions may apply to those who are concerned with the study of infection in the relation between parasite and host. In order to simplify the problems we are driven to regions in which certain diseases are just being introduced, because in such regions we may find the relation between parasite and host freer from the complexities which coëxistence for centuries has produced. Valuable material in this field is available in Africa, for example, where the incidence and morbidity of tuberculosis among African troops, exposed for the first time to Europeans, may be obtained. We know also that measles introduced into fresh aboriginal populations becomes a deciminating plague. The reference by Dr. Dochez to the manner of behavior of pneumonia in individuals hitherto unexposed to pneumococci belongs to the same class of phenomena.

Then there is the related problem of the interaction of primary and secondary agents of infection. Many instances of this kind are familiar to us all, as the pneumonia following measles, influenza, etc. This subject is one which we may penetrate experimentally. I wish to refer briefly to a single such instance: Two of the workers at the Rockefeller Institute, Drs. Olitzky and Gates, have made a study on rabbits of the influence of filtered extracts of the nasopharyngeal secretions of cases of epidemic influenza in the earliest stages. Such extracts, which are free from ordinary bacteria when injected intratracheally into the lungs of rabbits, injure the lungs slightly, producing punctiform hemorrhages and edema, and a leukopenia affecting the mononuclear cells. Animals thus treated recover in a few days, but if subjected to inoculation with minute sublethal doses of pneumococci, streptococci, staphylococci or Pfeiffer's bacilli, these organisms localizing in the lungs induce a pneumonic process to which the rabbit succumbs. Anaërobic culture methods have yielded a minute bacillus which acts in the manner of the filtered extract. We have here an instance in which two forces, one merely preparative and the other more injurious, work together to bring about a severe and fatal infection. Everyone will perceive in this experimentally devised combination of factors an analogy with the severe pneumonic affections attending epidemic influenza in man. Other examples could be given, but this one may suffice.

Dr. Emanuel Libman, New York: I should like to emphasize what Dr. Flexner stated regarding influenza. When Dr. Olitsky inoculated his organism into the trachea of rabbits he found interesting lesions in the lungs, consisting of edema, emphysema and hemorrhage. Dr. Loewe, independently, isolated the same organism and found the same lesions in infected animals. Dr. Baehr, in the course of pathologic studies, discovered that all these lesions are due to blood-platelet thrombi in arterioles and capillaries. The affinity of the organism seems to be for the bloodvessels and the hematopoietic organs. The lesion seems to be characteristic for the organism in question. In elaborate studies made by Baehr and Loewe similar lesions could not be produced by the usual bacteria nor by the virus or organism of epidemic encephalitis.

Dr. Theobald Smith, some time ago, promulgated the idea that during an epidemic many individuals were infected without showing any clinical evidence of disease. Studies made in typhus fever by Dr. Baehr confirm this view. In the course of the studies made in New York in 1914 it was found that complement-fixing bodies for the Plotz bacillus were absent in ninety-two controls and present in 87 per cent of the cases of typhus fever. In five of ten persons, however, who were in close contact with epidemic typhus fever, or who were doing experimental work on typhus fever (without full precautions), complement-fixing bodies were found. He therefore believed that persons coming in contact with typhus fever might become infected without giving any definite evidence of having the disease. During studies made in Russia in 1916 he confirmed these observations. There in a detention hospital he had the opportunity of studying persons who had been in contact with typhus fever. Some developed typhus fever and quite a large number of the others developed agglutination reactions in high dilutions against the bacillus of Plotz.

These observations are of importance whatever role the Plotz organism may play. In 192 control cases Plotz, Olitsky and Baehr found no agglutination reactions for this organism in a dilution of over 1 to 50; whereas, in typhus fever, agglutination reactions, often in very high dilutions, were found in over 90 per cent of the cases. Such serologic reactions do not, of course, necessarily indicate immunity, but they surely support the view of Dr. Theobald Smith.

THE ESSENTIAL IDENTITY OF THE ANTIBODIES

By HANS ZINSSER, M.D.
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Since Ehrlich's first classic analysis of antibodies it has been a generally accepted conception of immunity that agglutinins, precipitins, sensitizers, bacteriolysins, hemolysins or the so-called amboceptors, opsonins and the anaphylactic antibodies, are separate substances formed in the animal body, often in response to treatment with a single antigen. Krause, in the first edition of Kolle and Wassermann's *Handbook*, summarizes this point of view unambiguously in the following words: "Just as the bacterial body contains a variety of different antigens, so we may assume that animal protein is made up of a large number of different antigenic elements. If the animal body is treated with such substances and finds corresponding receptors there results the formation of a variety of qualitatively different antibodies. . . . When Gengou, in 1902, noted that alexin or complement was fixed when a precipitating antiserum was added to its homologous antigen, he interpreted this as meaning that, in addition to precipitins, the antiserum contained other antibodies—the "albuminolysins." To be sure, Gay and Moreschi showed that the fixation of alexin was chiefly a property of the precipitate which was formed, but this was regarded as signifying that the protein sensitizers were mechanically carried down during the precipitation. Ehrlich, commenting upon this in 1910, says: "It seems reasonable to assume, in accordance with Gengou's first explanation, that the property of binding the complement is exercised by the albuminous bodies sensitized with a specific amboceptor, and just as when immunizing with cells agglutinins and amboceptors are formed, so also when immunizing with dissolved albuminous

bodies two kinds of antibodies are formed, precipitins and amboceptors."

To be sure, the idea that agglutinins and precipitins might represent one and the same antibody has been more or less prevalent since their first observation. It originated, we believe, with Paltauf, and was expressed as a widely accepted view by v. Eisler in a review of precipitin reactions published in 1909. But in regard to identification of other antibodies with these two, there has been either a complete disregard of such a possibility or, when suggested, it has been thrown out of consideration because of the frequent and complete lack of quantitative parallelism between the curves of the various antibody functions in one and the same serum.

The idea of such a possible identity, however, has cropped up again and again, and in our own work has gradually forced itself upon us so insistently that we have thought it important to bring it forward again.

The fundamental idea was expressed quite definitely in 1908 by Bail and Hoke, who made an extensive study of the bacteriolytic, precipitating and agglutinating actions of normal beef serum and immune rabbit serum upon cholera spirilla. They clearly expressed the opinion that there were no separate bacteriolytic precipitating or agglutinating antibodies in those sera; that the essential fact was the existence, in the sera exerting these effects, of a single antibody which united with the bacterial substances, and that the various reactions which followed were the results of the conditions under which the different observations were made. Although a similar idea, on somewhat less valid evidence, had been expressed by Bürgi, and although Bail and Tsuda followed out the thought in subsequent publications, the view has made little actual headway in spite of much corroborative, though scattered, evidence.

In order that there may be no ambiguity as to just what is meant by what we call the "unitarian" view of antibodies, let us begin by formulating it clearly.

By such a conception of antibodies we do not, of course, imply that a complex cell like, for instance, the typhoid bacillus can give rise to one variety of antibody only. There may be formed a specific sensitizing antibody against the major chemical constituent

and other sensitizers against other antigenic substances enclosed in the same cell body or contained in the same antigenic solution. But we do mean that were we working with a single antigen, in a pure state, one variety of antibody only would be produced. This would be present in the form of a serum constituent specifically capable of uniting with the antigen. As a result of the union the antigen is altered in its physical and perhaps, to some extent, in its chemical behavior. The resultant reactions which may be observed with this sensitized antigen (agglutination, precipitation, complement-fixation, bactericidal phenomena, bacteriolysis, opsonization or sensitizing effects in the anaphylactic sense) would be determined not by differences in the nature of the antibodies with which the antigen had united but rather by the physical state of the antigen itself, the nature of the coöperative substances (alexin, leukocytes, tissue cells) and by the environmental conditions under which the observations are made.

Thus if the antibody comes in contact with a very finely divided antigen, as in a bacterial extract or in, let us say, horse serum, if electrolytes are present and perhaps other necessary physical factors furnished by the presence of serum, etc., precipitation occurs.

When we are dealing with whole bacteria of relatively large mass and correspondingly small surface exposure, agglutination is the result, and quantitative parallelism with the precipitin reaction is not to be expected because of the much greater dispersion of the antigen in the latter test.

When alexin is present, complement-fixation or hemolysis or bactericidal effects result, since the changes produced by the sensitization have now permitted union with the complement.

When there are leukocytes present the union makes possible the phagocytosis of the antigen, and when the antibody is absorbed by the cells of an animal, anaphylactic "sensitization" occurs.

As we have stated, the earlier opposition to such a view was largely based upon lack of quantitative parallelism between agglutination and precipitation curves on the one hand and bactericidal or protective antibody curves on the other, and this in spite of the relative inaccuracies which biologic measurements of this nature imply. In appraising such objections, however, we must not forget

that agglutination and precipitation are actually only secondary phenomena, after the union of antigen and antibody have taken place, and are dependent upon a great many environmental factors which may not, to the same degree, influence phenomena in which alexin, the leukocyte or the body cells of animals are involved. We need only to point out the frequently observed alterations and diminutions of the agglutination and precipitating powers of sera by heat. Heating antibacterial sera even to 56° to 60° C. will often materially diminish their precipitating effects for bacterial extracts, an observation which is entirely analogous to the influence of heating (to 60° to 70° C.) on the flocculating effects of serum for various colloidal suspensions, like arsenic trisulphide, etc. In addition to this the flocculation reactions depend upon the presence and the concentration of electrolytes, upon reaction, upon mutual relations of concentration, and perhaps upon viscosity. Moreover, the suspension equilibrium of the sensitized antigen must to some extent depend upon the varying factor of the inactive serum constituents carried into the union with the antibody, for we know that in precipitation reactions the bulk of the precipitate comes from the serum, and yet relatively protein-free antibodies can be split off from such a complex (Gay and Chickering, Chickering, Huntoon)—conditions which prove that, in the union, much inactive protein substance is carried along which inevitably must influence reactions of flocculation. Indeed, Tulloch has suggested that these "presumably inactive constituents" may even protect the united antigen-antibody complex from flocculation, a conception which we believe explains the "agglutinoid" phenomena.

It is not to be wondered at, therefore, that agglutination and precipitation curves should not run parallel with the curves of other antibody functions. And it is worth noting that in regard to such lack of parallelism, while it has frequently been noted that agglutinating and precipitating functions were often weaker than other antibody effects, or even absent entirely in such sera, it has rarely been observed that they were powerfully and specifically present when other effects were lacking.

In addition, however, to these purely quantitative objections to the "unitarian" conception other arguments have been advanced,

such, for instance, as those of Gay and Stone, who observed, with cholera extracts and cholera sera, that the formation of precipitates removed little or no lytic substance from the supernatant fluid. As against this, however, we have the experiments of Bail and Tsuda, who found that from specific cholera precipitates, obtained as above and injected into the peritoneal cavities of guinea-pigs, lytic and bactericidal antibodies were liberated and effective in producing a Pfeiffer reaction.

We must confess that such failure of quantitative removal of antibodies from the supernatant fluid, after agglutination and precipitation reactions had been carried out with these sera, was our own experience with typhoid serum, and there are certain points here that require further investigation. But when one considers, as we have stated above, that the amount of sensitizer necessary to bring about an agglutination or precipitation may be very slight, even in the presence of an excess of antigen, and that from such agglutinated or precipitated bacteria or bacterial extracts dissociation can take place; and if we then further consider the great inaccuracies coincident to the quantitative determination of bactericidal and bacteriolytic effects, such experiments lose much of their weight. One need only recall such experiments as those of Töpfer and Jaffe, who found absolute lack of uniformity between plating tests and the Pfeiffer phenomenon in determining bactericidal substances in one and the same sera. There are too many and uncertain secondary factors necessary in all these purely biologic methods to give one much confidence in any reasoning based purely on quantitative results.

More recently the growing interest in the purification of antibodies by dissociating them from antigen-antibody unions has again led to attempts to separate antibodies in the pure state. Landsteiner in his early work on dissociation makes no particular point of this phase of the problem. Gay and Chickering found that protective antibodies could be extracted from specific precipitates with dilute sodium carbonate, but interpreted this as a mechanical absorption of the protective bodies during precipitation. Huntoon succeeded in sensitizing pneumococci heavily, and then, by treating them with slightly alkaline salt solution at 55° C., dissociating from

them protective antibodies in considerable concentration and in solutions which are practically protein-free. With these antibody solutions he has been able to protect mice. It is not our function to go into the experiments of Huntoon more extensively, since these are all published or in the process of publication. The important point for us is the fact that these highly protective antibody solutions fail to agglutinate pneumococci in a large number of tests made by him, although they possessed distinct protective powers.

Having considered some of the more important objections, let us see what may be said in favor of the "unitarian" view apart from its simplicity.

The identification of agglutinins with precipitins should hardly require much argument, since both are specific flocculation reactions between a serum and the same antigenic substance, depending upon analogous environmental conditions. That they should differ from each other quantitatively is to be expected from the fact that in one case the antigen is present in relatively large masses and in the other case it is finely dispersed.

That there is at least a strong likelihood that precipitins and bactericidal and protective antibodies may be identical is indicated by the experiments of Bail and Tsuda, who dissociated bacteriolytic cholera antibodies from precipitates obtained with cholera extracts and normal beef serum.

Bail and Hoke obtained similar results not only with normal beef serum but with immune rabbit serum, though in the latter case somewhat less sharply. The latter point is important because the conglutination question is involved in their experiments with normal serum, for they obtained much better precipitates with the normal beef serum in its active state. However, since sensitization with an antibody is necessary for the conglutinin effect the principle of Bail and Hoke's experiment in regard to the liberation of bacteriolytic sensitizer from precipitated cholera extracts remains the same.

Also, the identity of precipitins and protective antibodies is very strongly suggested by the experiments of Gay and Chickering and of Chickering, inasmuch as they succeeded in extracting protective pneumococcus antibodies from specific precipitates, and the identity

of the two is at least as likely as mere mechanical carrying down with the precipitate.

The probable identity of precipitins with complement-fixing antibodies is indicated by the work of Dean and by many observations of our own.

In 1912 Dean, analyzing the relationship between alexin fixation and precipitation, concluded that the proportions of antigen and antibody, which favor rapid and complete precipitation, do not favor complete alexin fixation. He did not believe that the two reactions followed a parallel course, but said that he thought they represented two phases of the same reaction, a "Flocculation representing the first stage of a change that can be recognized in its early incomplete stage by complement-fixation." In 1912 and 1913 we were engaged in a similar analysis, from which we came to the conclusion that there was no need for assuming that the antibodies which were involved in the fixation of the alexin were essentially different from those that brought about the precipitation.

Studying the relationship between precipitin reaction and complement-fixation in the reactions between sheep serum and antisheep rabbit serum, like so many other observers we found that amounts of antibody which did not precipitate would, nevertheless, produce complement-fixation reactions in such mixtures, and this, of course, is the basic principle of the forensic complement-fixation introduced and described practically by Neisser and Sachs. In order to eliminate the possibility of non-specific mechanical carrying down of the complement-fixing antibody, we carried out experiments with complement fractionation and found that the fixation by the precipitates and by the supernatant fluids in such mixtures took place in the same way that fixation by sensitized cells occurs—namely, that the precipitates first fixed mid-piece, then end-piece secondarily. From this and other experiences we expressed the conclusion at that time that "There is but one variety of specific sensitizer," and that "The visible precipitation is merely secondary, occurring because of the colloidal nature of the reacting bodies under quantitative and environmental conditions which favor flocculation." Furthermore, the experiments of the last ten years have shown again and again that the physical state, the formation of

precipitates and the changes in sizes and perhaps in electric conditions of the substances in serum reactions are intimately related to the fixation of alexin. The recently developed observations on the Wassermann reaction, the Vernes test and the Sachs-Georgi reaction, now successfully employed in our laboratory, are all evidence in this direction.

That the anaphylactic passive sensitizing effect of sera was found to be quantitatively proportionate to the precipitin contents of these sera was shown by Doerr and Russ in experiments which seem to completely justify the identity of precipitating and anaphylactic antibodies suggested by Friedberger in 1908.

In regard to the dissociation experiments of Huntoon, some of whose solutions have been kindly sent to us by Dr. Huntoon himself, we have, indeed, found that these solutions exert considerable protective effect upon mice, and fail either entirely, or almost entirely, to produce agglutination *in vitro* of the pneumococcus against which they protect.

This would seem a potent argument against the "unitarian" idea. However, it must be considered that the "protein-free" antibody of Huntoon is physically, and therefore in its agglutinating and precipitating functions a quite different substance from the original antibody in the unchanged serum, which in its native state is inactive, associated with the pseudoglobulin substances which it carried down with it into precipitates. For, as we have stated before, it is, of course, known that most of the substance which comes down in precipitin reactions is derived from the immune serum. Moreover, by to some extent restoring the Huntoon antibodies to their original environment in the circulation of an animal we were able to observe powerful agglutination. We repeated, with these substances, the experiment on the mechanism of serum protection against pneumococci in rabbits carried out by Bull in 1915. A rabbit of approximately 1200 grams was intravenously injected at 5 P.M. with about 8 cc of a broth culture of pneumococcus I of relatively low virulence. At 10 A.M. the next morning the rabbit was very sick and a heart's blood puncture was done which showed numerous pneumococci evenly distributed through the blood stream. Ten cc of Huntoon material were injected intracardially in this

animal, then the needle was withdrawn and about two minutes later blood was taken from the heart and smeared. Similar punctures were done five minutes later and fifteen minutes later. Smears from the heart's blood taken two minutes later showed the pneumococci in the blood in clumps of varying sizes, with a very few small clumps of two and three and a very few individual organisms. But the large majority of the organisms were now in clumps of ten or more members. Smears after five minutes showed a few clumps and a few individual organisms, and after fifteen minutes there were very few single organisms and no clumps; but it is now difficult to find organisms, whereas before injection every field showed ten or more.

We did not succeed in thus restoring the agglutinative functions of Huntoon's materials in all cases, but the reason for this was revealed, we believe, by experiments carried out for us by Mrs. Parker. She found that the Huntoon substances produced by dissociating sensitized pneumococci in dilute sodium bicarbonate solutions at 55° C. had a reaction of Ph 8.8 to Ph 9.4. When powerfully agglutinating pneumococcus sera were subjected to similar treatment at similar reactions their agglutinating powers were largely lost and but partially restorable by neutralization. It would seem that these various considerations should serve materially to weaken the validity of objections to the "unitarian" theory based upon the separation of antibodies by dissociation experiments.

Furthermore, observations recently made by Coulter seem to us to have considerable bearing upon this question, indicating that agglutination and the sensitization to hemolysis of red cells are both due to the action of one substance. Coulter found that "The optimum hydrogen ion concentration for the agglutination of sensitized cells (rabbit antisheep sensitizer) in a salt-free medium occur at Ph 5.3, which corresponds with the optimal point for the precipitation of the serum-globulin itself." The optimum for precipitation of serum-globulin, in which the immune bodies are carried, is stated by Rona and Michaelis as Ph 5.2 and the iso-electric point for typhoid immune bodies as Ph 5.4. This reaction is more alkaline than that which is optimal for the agglutination of normal sheep

cells in saccharose solution, so that Coulter concludes, as far as agglutination is concerned, the behavior of sensitized cells is closely related to the properties of the immune serum. We quote directly from a personal communication from Coulter as follows: "Furthermore, an equilibrium has been found to exist between the amount of hemolytic sensitizer free and that combined with cells, the amount which combines at a pH of 5.3 being at the maximum and approximating 100 per cent." These observations indicate that the hydrogen ion concentration at which the agglutination of sensitized cells is most perfect corresponds with the iso-electric point of that part of the serum which makes the antibodies and also corresponds with the point at which the largest amount of hemolytic sensitizer is absorbed by the red cells. This to our mind would be strong evidence in favor of identity of the hemolytic sensitizer and the agglutinin.

We do not wish by any means to convey the impression that we consider the "unitarian" view as absolutely and rigidly proved. We do believe, however, that the denial of such a view necessitates the assumption that the injection of a pure antigen calls forth five or six fundamentally different reactions on the part of the tissue cells, a theory which would be justified only on the basis of incontrovertible proof.

Were there nothing further to be said in favor of the "unitarian" view one might at least wait for further evidence before one tried to deny an existing view, however complicated. But there is much to be said in favor of it, and evidence in this direction is accumulating.

We have believed in the probable truth of the "unitarian" view for a considerable number of years, with sufficient conviction to teach it as the most likely state of affairs. And while we cannot prove it in all the ramifications of the difficult experimental problems involved, we believe it has gone far enough certainly to throw the burden of proof upon those who still cling to the separation and the conception of separate structure for agglutinins, precipitins, bacteriolysins, etc.

REFERENCES

1. Krause: Kolle and Wasserman's Handb., 1st edition, iv, 617 and 618.
2. Gengou: Ann. de l'Inst. Pasteur, 1902, vol. xvi.
3. Gay: Centralbl. f. Bakteriol., orig., 1905, lxxxiii, 603.
4. Moveschi: Berl. klin. Wchnschr., 1905, vol. xxxvii; ibid., 1905, vol. iv.
5. Ehrlich: Collected Studies on Immunity, translated by Baldwin, 2d edition, p. 585.
6. Paltauf: Quoted from von Eisler, loc. cit.
7. Von Eisler: Kraus and Levaditi's Handb., ii, 835.
8. Bail and Hoke: Arch. f. Hyg., 1908, lxiv, 313.
9. Bail and Tsuda: Ztschr. f. Immunität., orig., 1909, i, 546.
10. Bürgi: Arch. f. Hyg., 1907, lxii, 239.
11. Gay and Chickering: Jour. Exper. Med., 1915, xxi, 389.
12. Chickering: Jour. Exper. Med., 1915, xxi, 248.
13. Gay and Stone: Jour. Immunol., 1916, i, 83.
14. Tulloch: Journal of the Royal Army Medical Corps, February, 1918.
15. Huntoon: Jour. Immunol., 1921, vi, 117.
16. Landsteiner: München. med. Wchnschr., 1902, vol. xlix, 1905.
17. Dean: Ztschr. f. Immunität., orig., 1912, xiii, 84.
18. Neisser and Sachs: Berl. klin. Wchnschr., 1905, xlii, 1388.
19. Zinsser: Jour. Exper. Med., 1912, xv, 529.
20. Friedberger: Ztschr. f. Immunität., orig., 1909, ii, 208.
21. Bull: Jour. Exper. Med., 1915, xxii, 466 and 484; 1916, xxiv, 25.
22. Doerr and Russ: Ztschr. f. Immunität., 1903, iii, 43.
23. Töpfer and Jaffe: Ztschr. f. Hyg., 1906, lii, 393.
24. Rona and Michaelis: Biochem. Ztschr., 1910, xxviii, 193.
25. Coulter: Jour. Gen. Physiol., 1921, iii, 309.

EXPERIMENTAL STUDIES OF THE PHARMACOLOGY OF QUINIDIN

By ALFRED E. COHN, M.D.
AND
ROBERT L. LEVY, M.D.

(ABSTRACT)

It has recently been shown that in certain patients suffering from fibrillation of the auricles the normal cardiac mechanism may be restored by the oral administration of quinidin sulphate. Knowledge concerning the pharmacological action of this drug is desirable for the clinic and is important in so far as it may furnish information concerning the nature of the fibrillatory process.

Experiments were done on twelve dogs. Anesthesia was accomplished by intratracheal etherization. The thorax was opened in the median line. A myocardiograph of the Roy and Adami type was sewed into the tip and the base of the ventricles and connected with a system of Marey tambours. Changes in the degree of cardiac contraction obtained by this method may be taken to represent alterations in the volume output. The blood-pressure was recorded from the right femoral artery; injections of the drug were made into the left femoral vein. In eight experiments the threshold for the production of transient auricular fibrillation was determined by faradization of the right auricle. Electrocardiograms were made at frequent intervals.

The results may be summarized as follows:

1. *Rate.* No constant effect was noted. There was acceleration four times, slowing four times and fluctuation in the remainder of the animals.

2. *P–R (conduction) Time.* The changes were slight and rela-

tively insignificant. There was prolongation four times and shortening once.

3. *T-wave.* A change in this portion of the electrocardiogram occurred eight times. This consisted either of reversal in the direction of the deflection or of increase in the voltage of the original wave.

4. *Blood-pressure.* In all of the animals a striking fall in blood-pressure was observed, the extent of the fall depending in a measure on the amount of drug injected. There was partial but never complete return to the former pressure level.

5. *Degree of Contraction.* There was always an increase in the height of the stroke recorded by the lever, the increase ranging from 16 to 162 per cent. A point was always reached after which further introduction of the drug caused diminution in volume output. The increase in cardiac contraction occurred simultaneously with the fall in blood-pressure, but persisted and often continued after partial restoration of the blood-pressure level. Unlike the augmentation accompanying the fall in pressure induced by histamin or hemorrhage the increase produced by quinidin is usually due to greater muscle-shortening and not to diastolic relaxation. The persistence of effect likewise indicates a direct stimulating action on the heart muscle. The relation of the fall of pressure to the height of contraction is, however, not finally determined by these experiments.

6. *Threshold for the Production of Auricular Fibrillation by Faradization.* In four experiments this was slightly raised; in four others there was no demonstrable change.

7. *Effect on Premature Contractions.* In one animal frequent spontaneous ventricular premature contractions were noted. These disappeared after the injection of quinidin.

8. *Mode of Death.* Only one animal died with ventricular fibrillation. In the others there was progressive slowing of the heart, sometimes with the occurrence of sino-auricular block. The auricles, as a rule, ceased before the ventricles. In the final curves were seen isolated, orderly ventricular beats.

9. *Lethal Dose.* The lethal dose (mg. per kg.) was extremely variable. In general the greater the fractionation of dosage the greater was the amount of drug necessary to cause death.

Experiments dealing with the effect of quinidin on the rate of conduction of the excitation wave through heart muscle are in progress.

DISCUSSION

Dr. A. D. Hirschfelder, Minneapolis, Minn.: I should like to ask if the speaker also tried atropin on the heart after the fibrillation had been stopped by quinidin. In 1908 I was able to show that some, but not all, cases of experimental fibrillation to faradic stimulation of the vagus were stopped from fibrillating for a time. We know that in the beneficial effect of slowing of the heart by digitalis it does not stop by fibrillation, which is largely a vagus effect and can be cut off sharply by atropin. In our laboratory we have cut it off by nicotin, which slows the rate of the ventricle without stopping the fibrillation, as a central and not a peripheral effect.

Dr. G. C. Robinson, Baltimore, Md.: The action of quinidin throws some new light on the problem of the cause of auricular fibrillation and the circumstances effecting fibrillation. It demonstrates the fact that the auricles may be affected by a chemical substance, and their tendency to fibrillate may be reduced thereby. I wish to recall an experience with auricular fibrillation which shows that the auricles may be influenced by a chemical substance in an opposite direction. Auricular fibrillation was observed in a patient brought to the hospital for hydrogen-sulphide poisoning. The fibrillation lasted two days and then ceased spontaneously, the normal cardiac rhythm returning. After the heart-beat had returned to normal there was no evidence of any cardiac lesion nor of any disturbance of the circulation. This observation was interpreted as auricular fibrillation induced by the exposure to hydrogen sulphide, and indicates that an anatomic lesion is not necessary for the production of auricular fibrillation. The action of quinidin indicates that a chemical substance can cause a cessation of fibrillation as well as bring about its onset.

Dr. A. E. Cohn (closing): To test for idiosyncrasy the dose of this drug clinically is from 0.2 to 0.4 gram. It is given at half-hour intervals, by mouth, in doses of 0.4 gram. The effect has usually been obtained by giving a total dosage of 1.2 grams.

Dr. Robert L. Levy, New York (closing): In three patients we have administered atropin after the normal rhythm had been restored by quinidin. In these individuals fibrillation did not reappear after the atropin injection.

A CASE OF ALKAPTONURIA, WITH A STUDY OF ITS METABOLISM*

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In the latter part of May, 1920, a male patient, aged forty-four years, entered the medical clinic with symptoms and signs of a midbrain lesion, possibly due to a tuberculoma. He was almost completely aphasic, and no history of his family antecedents or his previous medical condition could be elicited.

The first two routine examinations of the urine revealed a slight reduction with Fehling's solution, but no other abnormality was then noted. A day or two later, when happening into the laboratory, we were much impressed with the appearance of a specimen of the urine because of its peculiar mahogany-brown color. We were told that it belonged to our patient with the midbrain lesion and that it reduced Fehling's solution, which first appeared brownish black and later gave a reddish copper sediment. We naturally immediately suspected alkapton as the cause for the change. Our suspicions were further confirmed when it was found that the color of the freshly voided urine was the normal amber which gradually deepened to dark mahogany brown on standing. Further special tests of the urine confirmed the presence of alkapton and the patient was transferred to the metabolism unit for more careful study.

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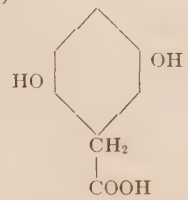
Before giving the results of this study let us briefly review the main points known about this anomaly of metabolism: Boedeker,[1] in 1861, first detected a reducing substance in the urine to which he gave the name "alkapton" (a bilingual word derived from alkali, and καπτειν, to absorb greedily), because of its behavior toward alkalies. Some years later (1891) Wolkow and Baumann[2] identified alkapton as homogentisic acid with an empirical formula of $C_8H_8O_4$. Later Baumann and Fraenkel,[3] as well as Osborne,[4] and Neubauer and Flatow,[5] successfully synthetized it and proved its structural formula to be that of paradioxyphenylacetic acid (hydroquinon-acetic acid).



This substance can be readily separated from the urine by the addition of solid neutral lead acetate, the resultant dense precipitate filtered off and the clear yellow filtrate allowed to stand in the cold, when the crystals of lead homogentisate appear, which later can be purified into colorless crystals of homogentisic acid, with a melting-point of 146° C.

Qualitative Tests. The urine on exposure to the air or upon the addition of alkali turns brown or even black. It reduces copper solutions but does not yield to the fermentation test, does not reduce alkaline bismuth solutions and does not rotate polarized light. An ammoniacal solution of silver nitrate is quickly reduced by it in the cold. Lastly when a dilute solution of ferric chloride is added to the urine, drop by drop, a deep blue color appears for a moment, as each drop falls, until oxidation is complete.

Incidence. It is a rare condition. Not more than 70 cases exist in the literature, and only 11 have been reported from America.

[1] Ann. d. chem. u. pharm., 1861, cxvii, 98.
[2] Ztschr. f. physiol. Chem. (Strassburg), 1891, xv, 228.
[3] Ibid., 1895, xx, 219.
[4] Jour. Physiol., 1903, xxix, 13.
[5] Ztschr. f. physiol. Chem. (Strassburg), 1907, lii, 375.

Barton Brune,[6] of Baltimore, reported in 1886 the first case in this country. Subsequently cases were published by Marshall,[7] of Philadelphia, Ogden,[8] of Milwaukee, and Osler,[9] of Baltimore, etc., making a total of 12 with ours. There is an unquestionable familial tendency, especially in the offspring of consanguineous marriages, according to A. E. Garrod.[10] Although familial it is not congenital, and no reports of parent and child being afflicted have ever appeared. Garrod's series of 11 cases occurred in four families, in all of which the parents were first cousins. In its incidence it shows a striking similarity to albinism. It is more common in males than in females.

Symptomatology. As a rule there is no inconvenience experienced by the patient except possibly the staining of the underclothing by the urine. Occasionally there is dysuria. It is often not recognized until the patient submits to a life-insurance examination and the applicant rejected as a diabetic. One of Osler's cases had been treated for mild glycosuria by four prominent continental physicians before consulting Osler. A more careful analysis of the urine by T. B. Futcher[11] revealed the fact that the reducing substance was alkapton. Though usually constant it may be intermittent and absent for years at a time. A small proportion of the cases (24 cases up to 1919) are associated with ochronosis or blackening of the cartilages and fibrous tissues and pigmentation of the skin. Alkaptonurics with an associated ochronosis may reveal during life pain, swelling and deformity of one or more of the larger joints of the extremities or of the small joints of the hands—a condition termed by Osler ochronotic arthritis. To explain this condition, Albrecht[12] considered that homogentisic acid or one of its derivatives combined with the chondromucoid or the chondroitin sulphuric acid of the cartilage to form the pigment.

In a few rare instances temporary alkaptonuria has been found in association with gastro-enteritis, gastric ulcer, pulmonary tuber-

[6] Boston Med. and Surg. Jour., 1886, cxv, 621; cxvi, 83.
[7] Med. News, Philadelphia, 1887, l, 35.
[8] Ztschr. f. physiol. Chem. (Strassburg), 1895, xx, 281.
[9] Lancet (London), 1904, i, 10.
[10] Inborn Errors of Metabolism, 1909 (London), p. 41.
[11] New York Med. Jour., 1898, lxvii, 67.
[12] Ztschr. f. Heilk., 1902, xxiii, 366.

culosis, peritoneal tuberculosis and diabetes. Falta and Langstein[13] have shown that some patients with grave diabetes have less power of destroying homogentisic acid administered by mouth than have normal persons.

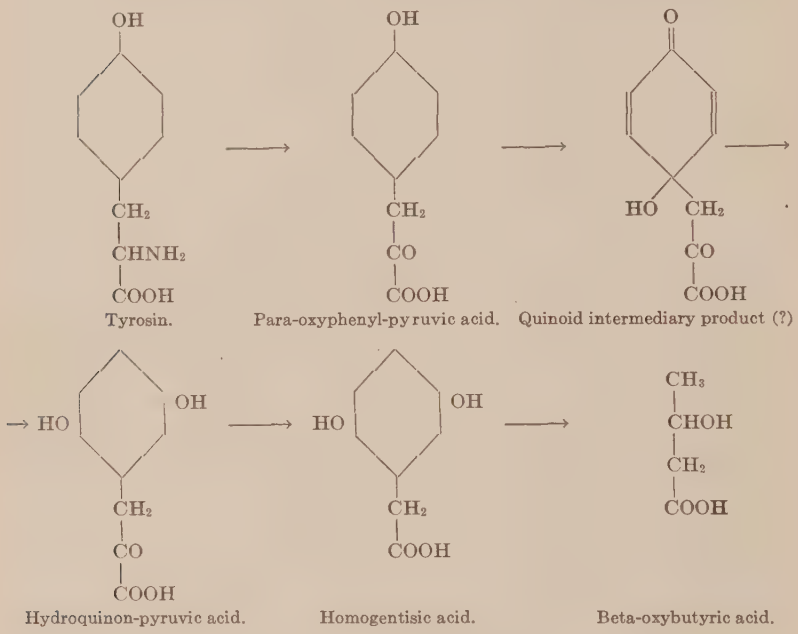
Pathogenesis. It is a freak of metabolism or, as Garrod so well expresses it, "an inborn error of metabolism." The error consists in a failure to complete the catabolism of the aromatic fractions of the proteins, tyrosin and phenylalanin. The tyrosin and phenylalanin of both the food and tissue proteins are concerned. During fasting the output of homogentisic acid is diminished but is not arrested. A diet rich in proteins greatly increases the output, and when tyrosin or phenylalanin is given by mouth to an alkaptonuric an almost corresponding quantity of homogentisic acid appears in the urine. On the other hand, these amino-acids when taken by normal persons are destroyed and the benzene ring broken up. The quantities of homogentisic acid excreted by different alkaptonurics are singularly uniform when the nature of the diet and ages of the patients are allowed for. This amount varies from 3 to 6 grams in twenty-four hours, although rarely 16 and even 18 grams have been found. Garrod states there is reason to believe that the error is in all instances complete and maximal in degree. Wolkow and Baumann[14] suggested that the change from tyrosin to alkapton might take place in the intestine under the influence of a rare specific microbe, but Garrod supports the view that the change occurs in the tissues after the absorption of the tyrosin rather than in the alimentary canal. Normally at least a part of the benzene ring may be broken up in the body, involving the production of the beta-oxybutyric acid; two of the carbon atoms of the latter are derived from the phenyl ring and two from the side chain, according to Wakeman and Dakin[15] (Fig. 1). Neither tyrosin nor phenylalanin yield glucose in the phlorhizin diabetic dog as do some of the non-ring form amino-acids (Lusk).[16]

In alkaptonurics the conversion of tyrosin and phenylalanin

[13] Ztschr. f. physiol. Chem. (Strassburg), 1903, xxxviii, 513.
[14] Loc. cit.
[15] Jour. Biol. Chem., 1911, ix, 139.
[16] The Science of Nutrition, third edition, 1917, p. 196 (W. B. Saunders Company, Philadelphia).

into homogentisic acid is so complete that the ratio of homogentisic acid to the total nitrogen in the urine tends to be constant and is the same for all cases (Garrod). The homogentisic acid amounts to 40 to 45 per cent of the total nitrogen figures.

FIG. 1.—Scheme of the origin of homogentisic acid (adapted from Neubauer).



Gross,[17] in 1914, found that the sera of normal animals and man destroy homogentisic acid, probably producing acetone; this change is brought about by an active enzyme which seems to be absent in the alkaptonuric. Since our own observations have been made, Katsch[18] has reported the case of boy, in whom he had earlier observed the disappearance of homogentisic acid from the urine during a starvation period. Subsequent study proved that the diminution of the alkapton condition was associated with ketone acidosis; at least the acidosis induced by a diet poor in carbohydrates reduced the H : N quotient to 1.5. The coincident acetone

[17] Biochem. Ztschr. (Berlin), 1914, lxi, 165.
[18] Deutsch. Arch. f. klin. Med. (Leipzig), 1918, cxxvii, 210; 1920, cxxxiv, 59.

elimination was greater than theoretically could be obtained from the aromatic amino-acids alone.

In order to prove metabolically the present case an alkaptonuric, the patient was put on a constant diet (70 grams protein, 83 grams fat and 253 grams carbohydrate; no meat or soup), the homogentisic acid and the total nitrogen excretion determined and the effects of tyrosin added to the diet observed. Homogentisic acid was determined by Garrod's modification of Baumann's method.

Day.	Total N, gms.	Homogentisic acid.	
		Grams.	H–N quotient.
June 11, 1920	6.27	3.77	60
June 12	7.84	4.78	61
June 13 (2 gm. tyrosin were ingested)	6.08	6.27	103

The homogentisic acid is higher in relation to the total nitrogen than is stated by Garrod, owing to the fact that milk was a principal constituent of the diet and casein contains more of the aromatic amino-acids than many of the common proteins. The characteristic increase in homogentisic acid is observed to follow the tyrosin ingestion. Subsequent determinations (Table I) show a quotient of 42 to 57, with an average for the five days of 50; at this time the patient was on a varying diet and the food intake was not controlled.

The distribution of the several nitrogenous constituents of the urine in alkaptonuria is said to remain normal (Lusk).[19] Only by high elimination of the homogentisic acid is the ammonia increased (Schumm).[20] Adler[21] states that purin metabolism is not disturbed. Ravold and Warren[22] found the urea below the average and the uric acid somewhat diminished. Interest in the sulphur metabolism was early directed to the possible conjugation of the homogentisic acid, but the acid was found to be free or combined as simple salts (Meyer).[23]

Certain difficulties are encountered in the determination of uric acid and creatinin by current methods in the presence of homo-

[19] Loc. cit.
[20] München. Med. Wchnschr., 1904, li, 1599.
[21] Biochem. Ztschr. (Berlin), 1909, xxi, 5.
[22] Jour. Biol. Chem. (Baltimore), 1910, vii, 465.
[23] Deutsch. Arch. f. klin. Med. (Leipzig), 1901, lxx, 443.

TABLE I.—NITROGEN PARTITION AND HOMOGENTISIC ACID EXCRETION

Day.	Volume, cc	Sp. gr.	Total N, gm.	Homogentisic acid.		Urea N.		Ammonia N.		Uric acid N.		Creatinin N.		Creatin N.		Undetermined N.	
				Gm.	Per cent. H/N.	Gm.	Per cent.	Gm.	Per cent.	Gm.	Per cent.	Gm.	Per cent.	Gm.	Per cent.	Gm.	Per cent.
June 18 . .	1040	1.030	11.76	6.08	51	...	...	...	...	...	...	0.423	3.60				
June 19 . .	2000	1.025	12.93	5.77	45	8.40	65.0	0.952	7.36	0.229	*1.77	0.398	3.08				
June 20 . .	1250	1.028	11.62	6.70	57	6.51	56.0	0.838	7.21	0.271	2.33	0.448	3.86				
June 22 . .	2000	1.018	15.00	6.27	42	8.46	56.4	1.120	7.46	0.233	1.55	0.435	2.90	0.085	0.56	4.68	31.2
June 23 . .	1340	1.032	13.86	7.58	55	7.24	52.2	1.160	8.35	0.189	1.35	0.438	3.16	0.064	0.46	4.77	34.4

* Morris's method (Morris, L. C.: Jour. Biol. Chem., 1919, xxxvii, 231); other determinations of uric acid are according to Folin and Wu.

SULPHUR PARTITION

Day.	Total SO_3., gm.	Inorganic SO_3.		Ethereal SO_3.		Neutral SO_3.	
		Gm.	Per cent.	Gm.	Per cent.	Gm.	Per cent.
June 22 . .	2.356	1.919	81.47	0.140	5.94	0.297	12.59
June 23 . .	3.218	2.520	78.33	0.319	9.90	0.379	11.77

gentisic acid. This is especially true of creatinin which was finally determined after the removal of the homogentisic acid with lead acetate. The silver lactate reagent of Folin and Wu[24] was promptly reduced in the cold, a reaction which should serve to identify homogentisic acid as other reducing substances that are found in the urine do not behave in this way.

In our case we found a high ammonia nitrogen, low urea nitrogen, moderately high uric acid nitrogen figures, creatinuria and high undetermined nitrogen. The sulphur excretion seems to have been essentially normal.

DISCUSSION

Dr. J. B. Futcher, Baltimore, Md.: Dr. Howard has mentioned the rarity of this condition and has spoken of the relationship of this metabolic disturbance to ochrinosis (which means ochre-colored cartilages). I have reported a case of alkaptonuria with ochrinosis in a patient in whom the cartilages of the ears and finger-joints and the cheeks had this peculiar slate color. There were three cases of ochronosis in his family. Osler used to speak of an ochronotic arthritis in the first patient, and there was distinct crepitation of the joints on palpation.

[24] Jour. Biol. Chem. (Baltimore), 1919, xxxvii, 459.

FAT–CARBOHYDRATE RATIO

By WILLIAM S. LADD, M.D.
(By Invitation)
AND
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As the treatment of diabetes mellitus is largely a dietetic problem, with the endeavor to render and maintain the urine free from glucose and the ketone bodies, to reduce the level of blood sugar to within normal limits and to supply a diet of sufficient caloric value to provide for the daily requirements, a thorough appreciation of the metabolism and interrelationships of the food constituents is necessary. To supply sufficient calories for maintenance in cases of severe diabetes it is necessary to employ large amounts of fat. Although the complete oxidation of fat requires that carbohydrate shall be simultaneously burned has been known for a long time, no very exact information as to the amount of carbohydrate necessary for the proper oxidation of fat was available until Zeller[1] reported the results of his experiments, in which he gave to two normal dogs and two normal men high calorie diets containing small amounts of nitrogen, with fat and carbohydrate varied isodynamically to demonstrate the level at which ketone bodies appeared in the urine. When the carbohydrate was reduced so as to constitute only 10 per cent of the caloric intake, ketone bodies appeared in the urine. Besides the practical bearing these experiments have in dietetics a matter of considerable fundamental importance is emphasized by Lusk,[2] namely, the possible molecular relation between carbo-

[1] Arch. f. Physiol., 1914, p. 213.
[2] The Science of Nutrition, 1917, third edition.

hydrate and fat in the complete oxidation of the latter. As Lusk points out, it appears "Possible that in the normal combustion of fats each molecule of B-oxybutyric acid, which is the end-product of the oxidation of each fatty acid, requires the presence of a triose molecule."

In view of the importance of the fat-carbohydrate ratio, from both the practical and scientific standpoint, we have made observations on individuals severely ill with diabetes who received diets that were devised somewhat along the lines of Zeller's experiments. Every case was rendered aglycosuric, aketonuric and the glucose tolerance determined. During the time of observation the following conditions were established, namely, the urine was free from sugar and ketone bodies, the diet was so constructed as to contain sufficient calories for maintenance, nitrogen equilibrium was secured (protein constituted approximately 10 per cent of the total caloric intake) and all variations in the fat and carbohydrate portions were made isodynamically. The various dietary levels were maintained for at least three days and in many instances five days. For the appearance of ketone bodies the qualitative test the sodium nitroprusside and ferric chloride reactions were used. In a few cases the ketone bodies in the urine were estimated quantitatively. In several of the experiments microscopic examinations of the stools revealed the fact that in all cases the food was well absorbed.

In the analysis of our results we have made use of certain more or less well-established facts in metabolism. Protein we have considered as yielding 58 per cent and fat (glycerol) 11.4 per cent of their energy in the form of carbohydrate. On this basis, provided the individual is in nitrogenous equilibrium, when the total available carbohydrate constitutes less than 10 per cent and fat more than 80 per cent of the total calories, ketone bodies in considerable amounts appear in the urine, indicating incomplete oxidation of the fatty acids in the organism. Shaffer[3] has reported similar observations. Our results furnish additional evidence of the probable molecular relationship between glucose and fat in the oxidation of the latter, that is, one triose or possibly two triose molecules are

[3] Proceedings of the American Society of Biological Chemists, Jour. Biol. Chem., 1921, xlvi, vi.

necessary for the proper combustion of one fatty acid molecule. It should be remembered that certain of the amino-acids (leucin, tyrosin and phenylalanin) produce B-oxybutyric acid, but the facts available in this connection do not admit of satisfactory quantitative estimations.

For practical purposes, then, in the construction of diets containing relatively large amounts of fat the ratio of carbohydrate available from the protein, together with the carbohydrate as such in grams to the fat in grams, should never exceed the ratio of 1 to 4. A more reasonable and conservative ratio is probably nearer 1 to 3. The latter ratio is quite adequate to supply the needs of any patient ill with diabetes who thus far has come under our observation.

OBJECTS AND METHOD OF DIET ADJUSTMENT IN DIABETES

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In the dietetic management of diabetes we are engaged in the effort to correlate symptoms and signs shown by the patient with the kinds and quantities of food that he consumes. The success of treatment, the average of results in all types of cases, depends on the truth of our concept of the relationship existing between symptoms or signs and the food supply. During the last few years the average of results obtained in the dietetic management of diabetes has been improved greatly through the work of Allen and Joslin, and the system they have developed is in some respects more logical and less empirical than any we have had heretofore. Yet the literature of the subject is still confused by a lack of unanimity among all writers as to the best manner of handling all cases. In a recent monograph, Falta[1] has again told the merits of his "cereal cure" and endorsed methods of management that differ materially from that which has found so much favor in this country. Newburgh and Marsh,[2] failing to achieve practical results by their application of the principles of "total dietary restriction" and "undernutrition," resort to low protein high fat diets with striking success in the management of seventy-four cases. In the past, good results were obtained and may still be obtained in some cases by old-fashioned "rigid" diets. The remarkable improvements that have sometimes been seen with the institution of a Donkin "milk cure," a

[1] Die Mehlfrüchtekur bei Diabetes Mellitus, Berlin, 1920.
[2] Arch. Int. Med., May, 1920, xxv, 647.

von Duering "rice cure," a Mossé "potato cure," a von Noorden "oatmeal cure" or any one of several analogous procedures cannot be denied and have never been explained fully to the extent that one may predict with certainty just when one of these empirical procedures will and when it will not produce a result better than that attainable by a more systematic method. The present study was made in the effort to correlate some of these varying views of different writers, and, if possible, to clear away some of the confusion that they tend to create in the mind of the physician as to what method of procedure he may best follow in any specific situation.

Discrepancies in the clinical literature of diabetes arise from three main sources: (1) Confusion in the minds of writers as to the exact nature of the anomaly of the metabolism which characterizes diabetes; (2) a general tendency among clinicians to think of the food supply of the body too exclusively in terms of the diet to the neglect of endogenous factors, and (3) the almost universal custom of thinking of the food supply simply as so much carbohydrate, so much protein, so much fat and so many calories without further analysis.

It will be necessary to develop each of these points separately.

1. Nature of the Diabetic Anomaly and Principles of Treatment. (*a*) One single specific defect characterizes diabetes. This consists in an inability on the part of the body to utilize as much glucose as may be utilized by the normal body when the supply of glucose exceeds certain limits. The diabetic appears to be capable of utilizing a limited quantity of glucose as well as the normal individual, but fails to utilize a normal percentage of any glucose introduced into the body in excess of this limit. This limitation of the body's power to utilize glucose is present in every case of diabetes. In its absence we cannot say that diabetes exists. In its presence alone, without any other accompaniment, one must say that diabetes does exist.

(*b*) The endocrine function of the pancreas, so far as we know it at all, is a single highly selective function having to do with the utilization of glucose and nothing else. All that we know of this function is deduced from our knowledge of the changes which occur in the diabetic metabolism and from the established relation between

diabetes and the pancreas. Were it not for the occurrence of the metabolic phenomenon which is called diabetes we would have no more reason to speak of a special endocrine function of the pancreas than we now have for speaking of such a function in the case of any of the organs which are not considered as endocrine organs in the usual sense of the word. No second metabolic disturbance besides diabetes has been found to occur with consistency after reduction of pancreatic substance. It is accordingly necessary to conclude that the work of the pancreas as an endocrine organ, so far as we know it today, has to do solely with the disposition of glucose, and that to lessen this work one must lessen the quantity of glucose entering the metabolism. The pancreas is a glandular organ innervated from the celiac plexus. Vagus and sympathetic fibers pass into its substance. It is a nerve-gland apparatus. Presumably it performs its function by secreting a substance having to do with the dissociation of the one specific sugar glucose into which all other carbohydrates that are capable of utilization on the large scale must first be converted before they can be oxidized, reduced or built up into glycogen. That such an apparatus should be stimulated by glucose to provide for the disposition of glucose is a thought in keeping with many facts. That a weakened pancreas in the presence of a sufficient supply of glucose might be stimulated into a state of fatigue and decreased function, and that a sufficient diminution of the glucose supply might then lessen the stimulation, place the organ in a state of comparative rest and permit it to recuperate up to a certain limit is also quite conceivable and affords a logical explanation of many observed facts.

(*c*) The anomaly of the metabolism in which abnormal quantities of acetone, acetoacetic and β-hydroxybutyric acids appear in the tissues, blood and urine is not due directly to any impairment of the endocrine function of the pancreas. It is a secondary effect in the nature of a disturbed metabolic balance resulting from the withdrawal of oxidizing glucose, as when one stone is withdrawn from an arch all the other members may settle to a new position. This anomaly is not peculiar to diabetes nor constantly associated with it. It occurs in other diseases. It may be made to appear in a normal subject by starvation or a diet containing too low a proportion of

carbohydrate and too high a proportion of fat, and when this is done it may be made to disappear again simply by the addition of more carbohydrate to the diet. It appears to be the immediate result of the oxidation of certain fatty acids in the absence of a sufficient proportion of "oxidizing" (dissociated) glucose.

There is for any given individual at any given time a definite ratio between the quantity of glucose oxidizing in the body and the maximum quantity of ketogenic fatty acids that can be oxidized in the same time without the appearance of abnormal amounts of the acetone bodies. In other words, the quantity of oxidizing glucose fixes an upper limit to the quantity of ketogenic fatty acid that can be completely oxidized at the same time. As to the absolute magnitude of this ratio, and the degrees of its variation in different individuals in health and disease, it is too early to make a final statement. Some years ago, on the basis of chemical studies by Ciamician and Silber, and test-tube experiments with acetoacetic acid, I suggested a certain type of reaction as the basis of "antiketogenesis" in which one molecule of acetoacetic acid would react with one molecule of an alcohol or glucose.[1] Zeller,[2] working with normal individuals on ample diets consisting of carbohydrate and fat with very low protein contents, shifted the proportions of fat and carbohydrate without changing the total calories and saw acetone appear when the ratio of carbohydrate calories fell below 10 per cent of the total, that is, when the ratio of fat to carbohydrate in the diet in grams was about 4 to 1. Recalculating Zeller's experiments, Lusk estimated the relative quantities of sugar and higher fatty acid that might have been oxidizing together in the body. Allowing for the formation of sugar from the glycerol of the fat, and of glucose from the protein catabolized, and for some glucose from glycogen, but not for ketogenic amino-acids from protein, Lusk suggested that possibly one triose molecule was necessary for the complete oxidation of one of higher fatty acid, that is, two molecules of higher fatty acid to one molecule of glucose, or 3 gm. to 1 gm. P. A. Shaffer[3] has worked with test-tube experiments and with

[1] Woodyatt, R. T.: Jour. Am. Med. Assn., December 19, 1910, lv, 2109.
[2] Arch. f. Physiol., 1914, p. 213.
[3] Proceedings of the American Society of Biological Chemistry (Fifteenth Annual Meeting, December, 1920, p. 6), Jour. Biol. Chem., March, 1921, xlvi, 98.

diabetic individuals in which he conducted metabolism studies, including observations on the respiratory quotient at the time acetone first appeared. Shaffer calculated the ketogenic acids of protein on the basis of the quantities of leucin, tyrosin and phenyl alanin found by analysis in 100 gm. of ox muscle protein by Osborne, assuming that each molecule of these known acetone formers may yield one molecule of acetoacetic acid or its equivalent. As a result of his work, Shaffer suggests that one molecule of glucose is necessary for the complete oxidation of one molecule of acetoacetic acid, or one molecule of any higher fatty or amino-acid that yields one molecule of acetoacetic acid (or equivalent). The molecular weight of glucose being 180, of oleic acid 284, of palmitic acid 256, and the average of the two acids 270, the ratio found by Shaffer if expressed in grams would be about 1.5 gm. higher fatty acid to 1 gm. glucose. Working with diabetic patients on maintenance diets under conditions that made it probable that the proportion of foodstuffs in the diets corresponded fairly with those actually catabolized in the body, and estimating the glucose and fatty acid as hereinafter indicated, we have also observed at the time acetone appeared ratios of 1.5 or a little above or below this figure with considerable frequency in harmony with the work of Shaffer. Accordingly, even though it may prove necessary to correct this figure as data accumulate, and for individual cases, it would seem that for clinical purposes one will make no gross error if it is assumed that the ratio of higher fatty acids to glucose, which if exceeded will lead to acidosis, is likely to be close to 1.5 to 1 (in gm.). This refers to the materials actually catabolized and to the diet only under the stated conditions.

Treatment. It follows from the foregoing that the rationale of dietetic management in diabetes is to bring the quantity of glucose entering the metabolism from all sources below the quantity that can be utilized without abnormal waste and to adjust the supply of fatty acids in relationship to the quantity of glucose so that in the mixture of foodstuffs oxidizing in the body the ratio of the ketogenic fatty acids to glucose shall not exceed limits compatible with freedom from ketonuria. When, as, and if, under these conditions of relative rest for the pancreas, the glucose using function

improves, then the food supply may be increased gradually in so far as this can be done without disturbing the above relations.

2. Endogenous Factors of Food Supply, Illogical Diet Restrictions. When a man fasts he does not, of course, cease to produce heat. Normal men during a fast on light exertion have been observed to produce from 29 to 30 calories per kg. of body weight daily. For a 50 kg. man this would mean 1500 calories per day. During the first four days of a fast, Cetti produced on the average 29 calories per kg. for a total of 1618 calories per day, and catabolized 85.88 gm. protein for 329.8 calories and 136.72 gm. fat for the remaining 1288 calories.[1] In a case studied by F. G. Benedict,[2] on the second day of fasting there were produced 1768 calories, or 29.9 calories per kg., and the individual was estimated to have catabolized 74.7 gm. protein, 147.5 gm. fat and 23.1 gm. glycogen. These figures are by no means the highest that might be selected. Thus, a well-nourished normal man weighing 50 kg., who during a fast produces 1500 calories per day may actually catabolize in the neighborhood of 75 gm. protein, 125 gm. fat and a little carbohydrate from glycogen. These well-known facts are repeated simply to emphasize the magnitude of the food supply from the tissues in fasting and to point out, in particular, that in fasting over 100 gm. fat may be thrown into the metabolic stream and catabolized daily. It has further been shown that the amount of fat in the fasting organism materially affects the amount of protein burned. In a critical review of the literature of the subject Lusk has said: "Where there was much fat present, little protein was consumed; when there was little fat, much protein burned; and when there was no fat, protein alone yielded the energy for life. In a normal individual the ingestion of fat will not prevent the death of the organism because there is a continual loss of tissue protein from the body which finally weakens some vital organ to such an extent that death takes place." But the ingestion of fat may spare tissue fat and thus prevent the protein loss from becoming abnormally great. It may be said that the ingestion of fat spares

[1] Citation from Lusk: Elements of the Science of Nutrition, third edition, 1917, pp. 86–89.

[2] The Influence of Inanition on Metabolism, 1907, p. 194, Table 128.

the individual any such protein loss as will occur if the tissue fat is allowed to become too much depleted. In this sense the ingestion of fat by an emaciated individual spares protein for that individual. Voit found in a fasting animal that the ingestion of suitable amounts of fat scarcely influenced the protein metabolism. To one dog "which in starvation burned 96 gm. fat Voit gave 100 gm. fat, with the result that it burned 97 gm. The fat ingested simply burned instead of the body fat, but the total amount of protein and fat burned remained the same" (Lusk).

Now, if a certain diabetic patient during a fast reacts essentially as a non-diabetic individual in the same state of nutrition, and if he weighs 50 kg. produces from 1250 to 1500 calories, and in doing so actually mobilizes and burns 100 to 120 gm. or more fat, the ingestion of an equal quantity of fat should leave his metabolism in the same state as before. The supply of fat would come at one time from the tissues, at another from the diet, but the quantity thrown into metabolism—the quantity presenting itself for disposition in the cells—would be the same in both cases. If these premises are sound, why then should we ever use complete fasting for diabetes? Granting that a diabetic patient may suffer from obesity as well as diabetics—that he may have two different metabolic defects, or granting that at times it might seem desirable to starve for other conditions besides diabetes, in such cases fasting would be rational if it would improve the general condition. But for diabetes itself, and particularly for diabetes associated with undernutrition, why for the purpose of desugarization should the patient be compelled to draw from his tissues the fat that he might draw from a diet, especially if in drawing from his tissues he lowers his fat reserves to the extent that he increases his protein losses? The striking results that have been obtained by Newburgh and Marsh with fat replacement diets bear significantly on this point. The practice of starving, or virtually starving, a patient in order to render his urine sugar-free, and then building up the diet, first with carbohydrate and then with protein, with a particular avoidance of fat, would appear to be based on the supposition that if fat were administered it would increase the catabolism of fat. But this would be in disregard of the endogenous food supply and illustrates

the necessity of thinking in terms of the metabolism rather than of the diet. As the diet falls the endogenous supply rises to take its place, and *vice versa*. The lower the diet the less its significance in calculating the food supply from all sources. It is possible to maintain the normal body with a diet that contains only 10 per cent more calories than are produced in fasting, and the differences in the catabolism of a man when receiving no diet and when receiving a 1500 calories diet may be slight.

3. Dealing with the Food Supply in Terms of Carbohydrate Protein and Fat. Carbohydrate, protein and fat are as such three separate and distinct substances, no one of which can be expressed quantitatively in terms of another, and if we speak of food supplies or diets as made up of so much carbohydrate, so much protein and so much fat we simply name them in terms of three variables. Thus each particular combination or diet becomes a specific, and having learned by experience how one of them will affect a certain patient, we have no means of knowing exactly how a second dissimilar combination will affect the same patient, much less another, except by trial and experience. The number of possible combinations of three variables is infinite and the number of practical food combinations, no two of which will differ by less than 5 gm. of one ingredient or by less than 50 calories, runs into the thousands. Accordingly, if we attempt to correlate symptoms and signs shown by the patient with the diet and follow the usual system of thinking of diets simply in terms of carbohydrate protein and fat without attempting to resolve them into simpler terms, it will be necessary to establish by experiment the effects of each diet combination in every type of case. It would seem tedious and hopeless to proceed by this inductive method. A further objection to this method, and a clear advantage in using another, lies in the fact that protein, carbohydrate and fat as such are not in reality the substances that present themselves for the final oxidative attack in the body which results in the liberation of energy. These substances are resolved by the processes of digestion and intermediary metabolism into simpler substances before they can be burned in the tissues. It is not starch in the bowel nor glycogen in the liver and muscles that taxes the endocrine function of the pancreas, but the glucose into

which these carbohydrates are resolved. Protein of the diet ceases to be protein and becomes a mixture of amino-acids before it can be absorbed from the bowel, and these undergo deaminations, etc., prior to actual oxidation. Neutral fats, to be sure, may be absorbed in part as such, and may be deposited in the tissues as such, but before they can be oxidized and used as sources of energy they must first be saponified into glycerol and higher fatty acids. Thus, as a matter of fact, carbohydrate protein and fat are not the actual foodstuffs with which we are dealing when it comes to the final metabolic processes. In the management of the diabetic food supply it is simpler and more rational to think in terms of the chemical metabolism. Falta devised a formula in which he added the carbohydrate of the diet to the urinary nitrogen times 2.8, or (following Lusk's suggestion) 3.65 to show the total quantity of glucose entering the body from carbohydrate and protein. Then, subtracting from this the quantity of glucose excreted in the urine, he obtained a figure for the quantity of glucose actually utilized. The quantity excreted divided by the total quantity supplied gives a fraction which multiplied by 100 was Falta's diabetic quotient. The latter has a limited value. The absolute quantity utilized is of more interest. The principle is important and can be developed further.

What Foods Become in the Body. Of the carbohydrates of the diet and tissues that play a significant role in heat production, all that do not already exist in the form of glucose are converted into this form by the processes of digestion or intermediate metabolism or both prior to ultimate utilization. We may therefore say that 100 gm. utilizable carbohydrate in the diet, if all is digested and absorbed, introduce into the metabolism about 100 gm. glucose.[1]

Fats of the diet in the course of digestion and intermediate metabolism must be saponified into glycerol and higher fatty acid before they can be oxidized. Fats such as tristearin, or an oil such as triolein, when completely saponified yield approximately ten parts by weight of glycerol to ninety parts by weight of higher fatty acid. Glycerol is capable of conversion into glucose in the body almost

[1] In tables of food analyses all that is listed as carbohydrate is not necessarily utilizable carbohydrate.

gram for gram. So we may say, for clinical purposes, that 100 gm. mixed fat in the diet, if completely absorbed and catabolized, will introduce into the metabolism about 10 gm. glucose and 90 gm. higher fatty acid.[1]

Protein of the diet, or tissues, is resolved into amino-acids, and in so far as these are absorbed and catabolized, they must be deaminized (and presumably at the same time oxidized) to yield oxy- or hydroxy acids. Of these a part is convertible into glucose, another part into β-hydroxybutyric and acetoacetic acids, while a third small fraction is destroyed in as yet unknown ways. According to the experiments of Lusk on phlorhizinized dogs, which have been repeated by others, and according to numerous observations in cases of maximal human diabetes, one may say that 100 gm. mixed food protein are capable of introducing into the metabolism approximately 58 gm. glucose. The same 100 gm. of protein also introduce a certain quantity of products which are quite the equivalent of products of the higher fatty acid catabolism in that they are capable of yielding β-hydroxy and acetoacetic acids. The exact quantity of these substances formed in the catabolism of 100 gm. protein can be estimated only roughly. The amino-acids that are certainly known to yield acetone bodies are leucin, tyrosin and phenyl alanin. If we take the quantities of these amino-acids found in 100 gm. ox muscle protein by Osborne and Mendel,[2] as Shaffer has also done, and convert the weights given into gram molecules, we obtain 0.16 gm. molecules of these ketogenic amino-acids. If we assume that each of these is capable of yielding one molecule of acetoacetic or β-hydroxybutyric acid and accept the view that one molecule of a higher fatty acid, such as oleic or palmitic acid, also yields one molecule of acetoacetic (or β-hydroxybutyric acid), then the 0.16 gm. molecule of ketogenic amino-acids would be equivalent in

[1] If objection is made to the view that neutral fats may yield 10 per cent of their weight in glucose on the grounds that ingested neutral fats have not been observed to cause the appearance of "extra" sugar in the urine of phlorhizinized dogs, it may be suggested that the ingestion of fat by a starving phlorhizinized dog would scarcely be expected to increase the fat catabolism for reasons already given unless special precaution had been taken to discharge tissue fat, etc. It is not improbable that the tissue fat that a fasting phlorhizinized dog catabolizes does produce glucose and that the glucose is then credited to protein.

[2] Jour. Biol. Chem., 1914, xvii, 366.

respect of its ability to form acetoacetic acid to 0.16 gm. molecule of higher fatty acid. If the acid were oleic (molecular weight 284) this would mean 45.44 gm. The nature of this estimation is such that considerable error is scarcely avoidable, but it affords a tentative figure that serves a practical purpose, even though it may require correction in the light of experience.

It will thus be seen that all of the foods of the diet, except a small fraction of the protein, resolve themselves in the body into two things: glucose, on the one hand, and higher fatty acid (or acetone forming equivalents) on the other. If we let G stand for the quantity of glucose and F A for the quantity of higher fatty acids that may be introduced into the metabolism by a mixture of carbohydrate, protein and fat, then—

100 gm. carbohydrate yields in the body .	100 gm. G and 0 gm. F A
100 gm. protein yields in the body . . .	58 gm. G and 46 gm. (?) F A
100 gm. fat yields in the body	10 gm. G and 90 gm. F A

These relationships may be expressed in the form of simple equations in which G is the total quantity of glucose introduced into the body by a given food combination; F A the total quantity of higher fatty acids (plus ketogenic amino-acids expressed in terms of higher fatty acid); C, carbohydrate; P, protein; and F, fat (neutral); thus

$$(1) \qquad G = C + 0.58\,P + 0.1\,F$$
$$(2) \qquad FA = 0.46\,P + 0.9\,F$$

If the ratio of F A: G, which if exceeded leads to acetonuria, is 1.5:1, then when F A:G = 1.5 we derive from (1) and (2) the equation F = 2C + .54 P, which for clinical purposes may be stated simply as

$$(3) \qquad F = 2C + \frac{P}{2}$$

Discussion of Hypothetic Diets. Let us now consider four hypothetic diets, I, II, III and IV and analyze them from the viewpoint of their possible effects on a certain diabetic patient weighing 50 kg. Let it be assumed that each diet is completely digested, absorbed and catabolized, and that each is exactly sufficient to cover the maintenance requirements of the body, so that we may in discussing them waive endogenous factors. Were these diets not

sufficient to maintain the body, foodstuffs derived from the tissues would add themselves to the combination actually being utilized in the body, and these would have to be known and allowed for. Also if some of the fat were deposited in the tissues this would have to be allowed for.

Looking at these diets simply as so many different combinations of carbohydrate, protein and fat, and going no further, they appear to be quite dissimilar. Who could certainly judge their relative effects on a patient? Diet I is a fair example of the old-fashioned "rigid" diet with almost no carbohydrate and the protein at 3 gm. per kg. of body weight. Diet II is a high carbohydrate, low protein diet. It is the kind of a combination that might possibly be given

TABLE 1.—FOUR HYPOTHETIC DIETS

	I.	II.	III.	IV.
Carbohydrate	10	77	60	51
Protein	150	30	85	70
Fat	84	108	91	125

in a "rice cure" or a "cereal cure." This diet would permit the patient to enjoy 385 gm.—almost a pint—of boiled rice or other cereal; or he could have 154 gm. white bread and butter—three slices—with each of his regular meals. Diet III appears to be intermediate between Diets I and II in all respects. It contains less carbohydrate than Diet II but more than Diet I, and the fat is less than in Diet II and more than in Diet I. It contains nothing that is not contained in higher quantities by one of the other diets. Diet IV resembles Diet III but might be suspected of having a higher caloric value because of its fat. We make a rapid estimation of calories and note that Diets I, II and III each represent 1400, while Diet IV shows about 1609 calories. On the basis of calories alone, if any of the diets were not to be tolerated Diet IV would be the one to suspect. Now, if in each diet we add up the grams of carbohydrate plus 0.58 of the grams of protein plus 0.1 of the grams of fat, we find that Diets I and II are alike in that each is capable of introducing about 105 gm. glucose into the metabolism. They are not only isocaloric, but will yield the same quantities of glucose. If we assume that the patient is capable of utilizing from 105 to 110

gm. glucose per day, he might very well tolerate either of these diets. Of the two, he would be less likely to tolerate Diet I. Coming to Diet III, and estimating the glucose equivalent G, we found it to be 118. This harmless looking isocaloric diet will surely cause glycosuria in the patient who is capable of utilizing only from 105 to 110 gm. Diet IV, with 200 more calories than any of the other three, has the same glucose equivalent as Diets I and II and will probably be borne as well as either. The question arises in respect of Diet IV as to whether the high fat is permissible or in any sense objectionable. We estimate the higher fatty acid value of this diet F A by means of formula 2. Taking 0.46 the protein + 0.9 the fat we find 32 + 113 or 145 for F A. As stated, G is 104. Then the ratio F A: G = 1.4. As a general rule, we have found that acetone does not appear in the urine of an uncomplicated case of diabetes or remain permanently if present when the ratio does not exceed this, provided that the diet is completely absorbed and catabolized and sufficient for maintenance, so that endogenous factors of food supply do not complicate the calculation. Accordingly, we say that the quantity and proportion of fat in Diet IV are probably not too high for complete utilization in the normal manner and that there is probably nothing objectionable about it.

This leads us to the problem of calculating the theoretic optimal diet.

Estimation of Optimal Diets. If C = carbohydrate, P = protein, F = fat, G = glucose and F A = higher fatty acids (plus ketogenic amino-acids expressed in terms of higher fatty acid) we may say—as shown above—that the quantity of glucose which any given combination of foods may introduce into the metabolism is expressed by the equation: (1) $G = C + 0.58 + 0.1\,F$ and that the quantity of higher fatty acids (and equivalents) may be expressed as (2) $F\,A = 0.46\,P + 0.9\,F$. When the ratio F A: G exceeds a certain value, ketonuria develops. Assuming that this ratio is 1.5, then $\frac{C + 0.58\,P + 0.1\,F}{0.46\,P + 0.9\,F} = 1.5$, when the ratio of fatty acids to glucose is as high as it may be without ketonuria. Simplifying this we obtain $F = 2\,C + 0.54\,P$, or, simply, (3) $F = 2\,C + .5\,P$. If it

is agreed that the ratio F A : G shall not exceed 1.5 and that the relationships expressed in equations (1) and (2) are as given, then to estimate the optimal food combination or diet one may use equations (1) and (3). Given the quantity of glucose that the patient can utilize completely, assign this value to G in equation (1). Thus, if 100 gm. is the highest quantity of glucose derived from all sources that the patient can utilize, 100 gm. = C + 0.58 P + 0.1 F. In order to secure the maximal number of calories the diet must clearly contain every possible gram of fat (at 9 calories per gram) that the value of G and the relations expressed in (1) and (3) will permit, and consequently the lowest possible carbohydrate protein fraction (at 4 calories per gram). Also, as between carbohydrate and protein, the protein must be as low as possible and the carbohydrate as high as possible, for 1 gm. carbohydrate yielding 1 gm. glucose and 4 calories provides for the normal oxidation of 1.5 gm. of higher fatty acid. On the other hand, 1 gm. protein having the same caloric value as carbohydrate yields less glucose to support fat combustion, and besides this yields acetone itself. If the body weight of the patient be 50 kg. and 1 gm. protein per kg. is selected as a conservative minimum, then P becomes 50 gm. and the equation (3), F = 2 C + .5 P becomes F = 2 C + 25. We have already made G = 100 gm. Now, the glucose yielded by the 50 gm. protein will be 0.58 × 50, or 29 gm., leaving 100 — 29, or 71 gm., to be distributed between carbohydrate and fat. In other words, C + 0.1 F = 71. From this we obtain F = 710 — 10 C. But we also have from the above, F = 2 C + 25. So 2 C + 25 = 710 — 10 C, solving which C = 57 gm. (57.08). Substituting this value for C in F = 2 C + 25 we find F = 139 gm. (139.16). Then the optimal food combination that will fulfill the conditions and relations specified is: carbohydrate, 57 gm.; protein, 50 gm.; fat, 139 gm.; calories, 1680. Proving this diet we find the total glucose equivalent G = 57.08 + (0.58 × 50) + (0.1 × 139.1) = 99.99 as called for. Also F A = (0.46 × 50) + (0.9 × 139.1) = 148.19 and F A: G = 1.5 as required.

It is apparent that any addition of any foodstuff to this diet will make G greater than 100. If, on the other hand, one added more fat—say 10 gm.—and subtracted 1 gm. carbohydrate, G would

remain 100 and the calories would be increased by 86, but this would make F A: G greater than 1.5. The effect of changing the protein can be seen by comparing the caloric value of a series of optimal food combinations with the protein rising from 0 to 2 gm. per kg. (See Table 2.)

It will be observed that for each gram of glucose that can be utilized in the body there are 18 calories in the optimal food combination as here calculated. Each gram of protein in the food supply subtracts 3 calories from the optimal number. For mental calculation, if one knows that a patient can actually utilize a certain number of grams of glucose, take this number seventeen times as the approximate number of calories that he will be capable of using at best without glycosuria or acetonuria if the diet is most favorably balanced. Then knowing the patient's weight the severity or mildness of the situation becomes apparent.

TABLE 2.—SHOWING OPTIMAL FOOD COMBINATIONS WHEN G = 100 GM. (IN THE EQUATION G = C × .58 + .1 F); WHEN FA: G = 1.5; AND WHEN THE PROTEIN IS 0, 25, 50, 75 and 100 GM. (I.E., 0; 1.0; 1.5; AND 2.0 GM. PER KG. FOR A BODY WEIGHT OF 50 KG.)

	P.	C.	F.	Calories.	Difference in calories.
1	10.000[1]	83.333	166.666	1833.327	76.25 (2)-(1)
2	25.000	70.208	152.916	1757.076	76.25 (3)-(2)
3	50.000	57.083	139.166	1680.826	76.25 (4)-(3)
4	75.000	43.958	125.416	1604.576	76.25 (5)-(4)
5	100.000	30.833	111.666	1528.331	

REPORT OF CASE. To illustrate practical aspects of the principles discussed the following case will be found of interest:

A Greek, aged twenty-six years, was admitted to the hospital as an emergency case. He was very weak, languid and emaciated; body weight, 45 kg. There was a flush on the face, and the respirations were slightly increased when quiet in bed. The plasma carbon dioxide was 23.9 volumes per cent by Van Slyke. The breath smelled of acetone and the urine contained large quantities of sugar and acetone bodies. He had the

[1] No. 1 is hypothetic and could only be considered as the non-protein fraction of a larger combination.

symptoms and signs which are commonly taken to indicate a very severe case of diabetes.

First Stage. He was put to bed with artificial warmth and placed on a diet of 400 gm. of 5 per cent vegetables with clear broth and plenty of water. But after four days he still passed 32 gm. sugar in the urine with acetone and 2.3 gm. ammonia. His temperature on the fourth day ranged from 96.4° to 97.6° F.; the pulse from 56 to 60; and he was so weak that it was not deemed safe to continue the effort to desugarize at that time. This ended the first stage of management.

Second Stage. He was then given for five days a diet containing 114 gm. carbohydrate, 45 gm. protein, 15 gm. fat and 760 calories. G for this diet is 142 gm. During these five days he excreted on the average 85 gm. glucose per day, so that he was disposing of about 67 gm. glucose per day from the diet alone. His general condition improved and he was then desugarized by two days of complete fasting. Having remained sugar-free for one day he was then given 400 gm. vegetables of the group that contains not over 5 per cent of carbohydrate plus 1 liter of clear broth. This diet contained 12 gm. carbohydrate, 14 gm. protein and 104 calories, and G is 20 gm. But he promptly showed sugar in the urine and was again desugarized by one day of fasting. The patient had just manifested his ability actually to utilize at least 67 gm. glucose, yet the mere feeding of a diet that could yield but 20 gm. of glucose in the body induced glycosuria.

Third Stage. The urine was now free of abnormal quantities of sugar and acetone, and the patient was given a liter of broth and 200 gm. of fresh vegetables, which he bore without glycosuria. Then he received 400 gm. greens, then 1 egg, and so on until the diet contained 314 calories, when the urine again showed sugar. Again his condition was such that it was unsafe to continue these tactics in the effort to maintain him in the sugar free state. Three weeks had passed and three successive attempts had failed, although at no time had the diet been capable of introducing into the body as much glucose as the patient had actually utilized on the 760 calories diet of the second stage.

Fourth Stage. He was given a diet that contained 92 gm. carbohydrate, 103 gm. protein, 70 gm. fat and 1416 calories. G for this diet is 159. He was on this diet for two weeks and excreted on the average 50 gm. glucose per day. Thus, he was actually utilizing about 109 gm. glucose per day. His condition was much improved and he was then desugarized by two days of fasting.

Fifth Stage. He was now in the sixth week of management, free of glycosuria and acetonuria after his third desugarization, and the attempt was again made to build up his diet gradually. But at 780 calories glycosuria recurred. A fourth desugarization by fasting was followed by another attempt, but glycosuria developed with a diet of 1000 calories. He was desugarized a fifth time and again broke over at 1000 calories. Nine

weeks had elapsed. He was then established on a diet of 953 calories, with which he remained sugar-free. The diet now contained 43 gm. carbohydrate, 69 gm. protein and 56 gm. fat, with G = 88. The plasma carbon dioxide was 60 volumes per cent. There was no abnormal quantity of acetone in the urine and the urinary sugar as measured by the method of Benedict, Osterberg showed 454, 515, 486, 425, 494 and 400 mg. respectively per day on six successive days (tenth week).

At 22 calories per kg., even with a subnormal basal rate, this was barely a maintenance diet and it was apparent that unless something better could be done the outlook for the patient was hopeless. Much had been written about the harmful influence of too much protein in the diabetic diet. Naunyn, von Noorden, Falta and others have emphasized the necessity of keeping the protein of the diet within limits. A new interest had been given this subject by the recent report of Newburgh and Marsh. We decided to test this idea to its limit. So the patient was placed as nearly as might be in a state of specific protein hunger.

Sixth Stage. The diet was made up exclusively of rice and butter to contain 24 gm. carbohydrate, 2.5 gm. protein, 102 gm. fat and 1024 calories. The calories were therefore a little higher than he had yet been able to receive without glycosuria. He was placed on this diet and remained sugar-free with excretions of 305, 400, 370 and 392 mg. respectively on the four following days. There was no acetonuria. Now, the diet was rapidly increased: first by additions of rice alone, then by butter and rice together. In ten days it contained 84 gm. carbohydrate, 11.5 gm. protein, 162 gm. fat and 1837 calories. The urine at no time contained acetone and during these ten days had contained 308, 359, 384, 317, 545, 335, 439, 330, 486 and 333 mg. respectively of sugar. But the patient was not in nitrogen equilibrium. The urinary nitrogen was extremely low, yet it alone was exceeding that of the diet by about 1 gm. daily. So two eggs were added to the diet, which made it contain carbohydrate 84 gm., protein 25 gm. (or 0.6 gm. per kg.), fat 174 gm. and calories 2000. On this (if the stools were neglected) the patient came into nitrogen balance.

Thus, after nine weeks of trial the ordinary practice of undernutrition had failed to establish the patient in the non-diabetic state with more than 953 calories, whereas after two weeks of the rice-butter diet the calories had more than doubled and the carbohydrate had been increased to 60 per cent. This seemed like a miracle.

The 84 gm. carbohydrate when served in the form of boiled rice made a mass of about 400 gm., which was all that the patient would eat. The contrast between the trays that now came to him and those that he had formerly had aroused many remarks in the ward. Surely, it might seem that there were merits in protein deprivation. This is the type of result that astonished von Noorden when he first used his oatmeal cure. It is the type of result that has made the reputation of the von Duering rice "cure," the Mossé potato "cure," the Falta cereal "cures." However, if we examine this diet according to the equation and take the grams of carbohydrate plus 0.58 of the grams of protein plus 0.1 of the gram of fat we find that G is 116 gm., and it will be remembered that in the fifth stage the patient had shown his ability to utilize 109 gm. glucose, although at that time he had not remained sugar-free. He was not now doing much more. The question arose as to what would have happened had the high protein diet of the fifth stage had a value for G less than 116.

Seventh Stage. A diet was composed to contain 28 gm. carbohydrate, 118 gm. protein (2.7 gm. per kg. instead of 0.6 gm.), and 160 gm. fat. Total calories, 2024 (as against 2000). G for this diet is 112. The patient was given this diet and remained sugar-free and free of acetonuria. He remained on it for eight days, when it was increased to find his tolerance limit. This was found to be 119 to 120 gm.

Interpretation. Clearly, the merit of the rice-butter diet had not lain simply in its low content of protein. Four times as much protein had not been incompatible with a result apparently just as good. In retrospect it is clear that the patient had shown from the second stage on the inherent ability to utilize a goodly amount of glucose. His actual tolerance was probably higher on the last day than it was on the seventh, but as early as the fourth week he had burned almost as much glucose as he ever did later, so it cannot be held that the final result was simply made possible by a rising tolerance. Notwithstanding his manifest ability to burn glucose, he had repeatedly shown glycosuria on very low diets that were incapable of introducing into his body as much glucose as he was able to burn, paradoxical as it may seem. This tends to encourage

speculation. Shall we say that, because of the general emaciation, the starved kidneys had become abnormally permeable to sugar, that the "renal threshold for sugar" was lowered, introducing an element of "kidney diabetes"? Or shall we evade the issue and say that in such cases the attempt should not be made to maintain the patient in the non-diabetic state; that such patients should be fed and allowed to run sugar until they are stronger, in spite of all that we know of the power of an excess of glucose to diminish tolerance? Shall we entertain the idea that a diabetic may burn more sugar if he is given an excess than he can if he is given just the quantity that he can utilize without glycosuria? I should say No; these ideas are based on misconceptions of the food supply of the diabetic. It was clear in this case, from the events that followed, that the patient could be given every gram of glucose that he was capable of burning under any circumstances and still be kept from showing glycosuria, provided his food supply were suitably adjusted.

Earlier in this paper emphasis was laid on the necessity of conceiving the food supply of the diabetic patient as coming from the tissue as well as from the diet, especially when the diet falls below the maintenance requirements of the body. In the present case we are dealing with such a situation. All of the low diets that aroused wonder, because they induced glycosuria, even though incapable of introducing into the body as much glucose as the body could burn, were diets that failed to meet maintenance requirements. Consequently, when these diets were being used the patient was drawing a greater or smaller part of his food supply from his tissues. It is not only necessary in general to consider the supply of food from the tissues, but to consider the kind and quantities of food that will be supplied by the tissues of the particular case in hand. When a patient is thrown back on his own tissues for food he can only draw on the materials that are there. As emphasized by the quotation from Lusk, when in fasting the body contains much fat, much fat burns and little protein; when there is little fat in the body, little fat burns and much protein; when there is no fat, protein alone burns. But whatever the composition of the body, something burns while life persists. This patient had very little fat in his body, and the quantity of glycogen was certainly negligible. Therefore,

when this patient fasted he must have produced much of his heat from protein. He weighed 43 kg. If in fasting he had produced only 15 calories per kg., this would have totaled 645 calories, an amount of energy contained in 161 gm. protein. If he catabolized this weight of protein he would liberate 58 per cent of this weight, or 93 gm. glucose. Then if he had at this time the ability to burn 109 gm. glucose; the addition of 20 gm. from the diet would probably cause glycosuria. Unfortunately, in this case the basal metabolic rate and the urinary nitrogen were not actually determined during the fasting periods. But the literature contains many references to the "azoturia" of emaciated diabetics, and there can be no doubt that in such a case as this fasting induces a marked protein loss. Thus, fasting in the case of a sufficiently emaciated patient is the equivalent of a pure protein ration. In another case, with plenty of fat in the body, it is quite different as much tissue fat will burn and much less protein. It was pointed out earlier that the ingestion of fat by a fasting individual who already has plenty of fat in his body has very little effect on the protein metabolism. If too much fat is ingested it may increase acidosis and increase the protein loss, but in the right quantities it simply replaces tissue fat. But if the body fat is already depleted, and if, as a result of this, the protein breakdown has already become excessive, then the ingestion of fat will retard the prevailing protein breakdown and reduce it to a lower level. Thus, in the emaciated individual the ingestion of fat spares protein. In this diabetic patient, in this particular state, the feeding of enough fat should reduce the protein catabolism and thereby reduce the quantity of glucose thrown into circulation. When he was given the rice-butter diet he received at once 102 gm. fat for 918 calories, almost enough to replace his minimal fasting caloric requirements, and this put a rapid quietus on the excessive protein breakdown. The quantity of glucose from protein having thus been cut down, there was then opportunity to introduce more carbohydrate in the diet, which further suppressed the protein breakdown. The principal reason why this diet operated so well was because of its content of fat and because the body was so depleted in fat at the time when it was given. The patient was in a state of extreme fat starvation. When in the sixth stage of

management the patient did equally well on a low carbohydrate high protein diet, this was because the combined carbohydrate and protein of that diet introduced into the body no more glucose than had formerly come from the combined carbohydrate and protein of the rice-butter-egg combination, and because this diet also contained enough fat to prevent endogenous factors of food supply from coming into action. Thus, it will be seen how the administration of a diet in excess of maintenance requirements may under certain conditions actually reduce the quantity of glucose in circulation in the body. Were the fat dropped out of these last two diets more glucose would be thrown into metabolism by the increased breakdown of tissue protein induced by the lowering of the diet. This, I take it, is a possible explanation of a certain type of glycosuria which develops with prolonged undernutrition. I have seen a patient with diabetes similar to this one who could not be desugarized at all by fasting, but who cleared promptly when enough fat had been added to the diet with no other changes. This man, in fasting, catabolized so much protein that his protein sugar overtaxed this tolerance limit.

DISCUSSION

Dr. L. H. Newburgh, Ann Arbor, Mich.: This is another step away from the starvation diet. From the clinical point of view I believe that it is wrong to subject the diabetic to the prolonged effect of undernutrition. I have accordingly tried to devise a diet which would not produce undernutrition and would at the same time prevent glycosuria and acidosis. We found we could do this by using a diet whose chief constituent is fat. Dr. Woodyatt, working from another angle, has arrived at the same conclusion. He has worked out the optimal diet on the basis of the laws of metabolism, keeping in mind the fact that metabolism does not cease in starvation and that the use of body tissue for energy may be attended by the presence of more glucose in the blood than is the case when properly adjusted diets are fed. These ideas have led him to the use of a diet high in fat. Treatment will be advanced as soon as it is generally recognized that this is the rational basis upon which to build up a diet.

Dr. E. P. Joslin, Boston, Mass.: There is more in Dr. Woodyatt's paper than one can appreciate today. Already I have spent several hours

upon it and am much impressed with it. What Dr. Allen did for Guelpa's work I think Dr. Woodyatt has done for the work of Newburgh and Marsh. Before undernutrition was inaugurated diabetic children lived about one year. That was prior to 1914. The next series treated with undernutrition have lived over two years, and now children have an expectancy of over three years. Whereas before 1914 the average duration of life of diabetic adults was three years it is now five years, and if we include living cases it is nearer seven years. Therefore, as the result of the 1914 methods, marked extension of life of diabetic cases has actually taken place. Today I am hopeful that we can look forward to another advance by utilizing some of the ideas brought out by Drs. Newburgh and Marsh and elaborated and expanded by Dr. Woodyatt. What should be our attitude to these new ideas? Should one go over to the new method of treatment and increase the quantity of fat in the diet? Dr. Newburgh's cases became sugar- and acetone-free in the treatment. I have tried this treatment in the worst possible cases of diabetes who were not doing well under my care. As a result, in four cases none became sugar-free and none became free from acidosis. In one acidosis became so severe that coma threatened. The alveolar air had dropped to 14 mm. mercury. That does not mean that Dr. Newburgh's treatment should not be utilized. We can use it in selected cases. These four severe diabetics of mine felt that they deserved to know the results in Dr. Newburgh's 69 cases, and I asked him for the details. He kindly reported that of his original 69 cases 22 were traced, and of these 8 were dead. The condition of 47 cases was unknown. In the light of the new data so far available with this new method the question now arises, what shall I do with two boys with diabetes beginning at the age of twelve, but now at the end of three or four years of treatment by the old method? These boys show a carbohydrate tolerance of about 100 grams, protein 70 grams, and fat 65 grams. Shall I put these boys on lower protein and push up their fat? or shall I continue by those methods which have helped them for these three years? With these cases I shall not adopt new methods. We must be positive not to lose the advantages gained during six years.

Dr. F. M. Allen, New York: The production of acetone bodies involves not only the two factors of fat and carbohydrate but also a third factor, namely, the living organism. If the fat and carbohydrate are made available in some fixed ratio there is still the possibility that the organism may vary in its disposal of them under different conditions.

One such condition may perhaps be found in the level of the total nutrition. It is generally believed that a strict protein-fat diet causes acidosis in the human subject, which seems to be most marked with luxus nutrition. In the extremely emaciated diabetic, maintained in equilibrium on a thousand calories or less, acidosis may be absent or trivial on a diet without

preformed carbohydrate. Direct experimental evidence will be necessary to establish whether the relations between fat and carbohydrate are the same and whether the carbohydrate moiety of protein is equally efficient in preventing acidosis at high and at low levels of nutrition.

A second possible variable is the idiosyncrasy of different normal individuals. This was apparently illustrated in the experiments of Higgins, Peabody and Fitz, when they took the same diets and metabolized them similarly, as indicated by respiration tests, yet their production of acetone bodies differed widely.

A third variable on the part of the organism is age and a fourth is species. It is well known that a dog excretes no appreciable quantities of acetone on fasting or on diets which produce acetone in the human. Children are supposed to be more subject to acidosis than grown persons, and a very positive and striking difference exists between the young puppy and the adult dog, inasmuch as the fasting puppy develops acidosis fully comparable to that of the human and sometimes actually reaching a fatal degree.

Observations under certain fixed conditions therefore warrant conclusions only for those conditions, and a much wider scope of experiments must be necessary to prove that the same proportions of fat and carbohydrate supplied to the normal organism or cell must always be metabolized in the same manner. Still further possibilities of variation are offered by pathologic conditions. I have performed some animal experiments which I have not yet succeeded in publishing, but which seem to show a special influence of certain organs upon the production of acetone.

In regard to diabetic treatment animal experiments already published seem to me to show that the total caloric intake and the total body weight somehow constitute a load upon the internal secretory function of the pancreas. Contrary to my belief and wish, most of the diabetic patients in the Rockefeller Institute Hospital were given high calory diets and all the severe cases thus treated ended fatally. The only two juvenile patients who are alive today are the two who escaped this treatment and the sole known difference between them and the others is that their diets were kept relatively low in fat and calories. One of these two has lost tolerance seriously through surreptitious taking of fat, especially butter, and this decline of assimilation seems to have been halted by eliminating this error. The other child has maintained good health and high tolerance, in spite of having passed through a series of severe infections which might have been expected to injure the assimilation. Other clinical experience convinces me firmly that the attempt to give a high caloric diet or to build up the body weight too high with fat or any other food is injurious and leads to a fatal result in every genuinely severe case of diabetes.

Dr. Solomon Strouse, Chicago, Ill.: The excellent results reported in the last few years in the treatment of diabetes are due as much to the

care and skill of the physician as to any particular form of diet. There are two classes of diabetic patients who do not respond well to undernutrition: those who despite rigid adherence to a low diet do not maintain tolerance, and the other group is that of the older chronic diabetics who when made sugar-free at the expense of nutrition are more liable to develop symptoms. On the other hand, some patients get along very well on a higher fat diet even though they may have previously shown signs of acidosis. It is easier for a patient to live on a diet of 1800 calories than on one of 1000 calories. There is a difference between malnutrition and a luxus diet: there is the moderate diet which contains more fat than we have been accustomed to use.

Dr. W. W. Palmer, Baltimore, Md.: Dr. Allen says patients vary in their response to fat-feeding. In our experience, in the cases which we have considered the evidence satisfactory, as the diet is arranged so that the amount of carbohydrate constitutes about 10 per cent and the fat 80 per cent of the caloric value, there may be a temporary rise in the amount of ketone bodies excreted. When the carbohydrate fraction is made still less the amount of ketone bodies increases and remains more or less constant. It must be remembered that in the fat of the diet there is a definite amount in glycerol fraction which may be converted into glucose. Dr. Woodyatt has pointed out certain fundamental facts of metabolism which are of great service in the treatment of cases of diabetes.

Dr. R. T. Woodyatt (closing): Dr. Joslin's comments are sound. Every individual should not be put on the optimal diet. That should be left to the judgment of the clinician. When nutrition is the paramount consideration we must administer every possible calorie, and this method may help us to do so. Since beginning to treat cases in accordance with the principles described I have found no necessity for complete fasting in any. A case that can be desugarized by fasting can be desugarized with a fat replacement if G is made low enough.

FAMILIAL DIABETES

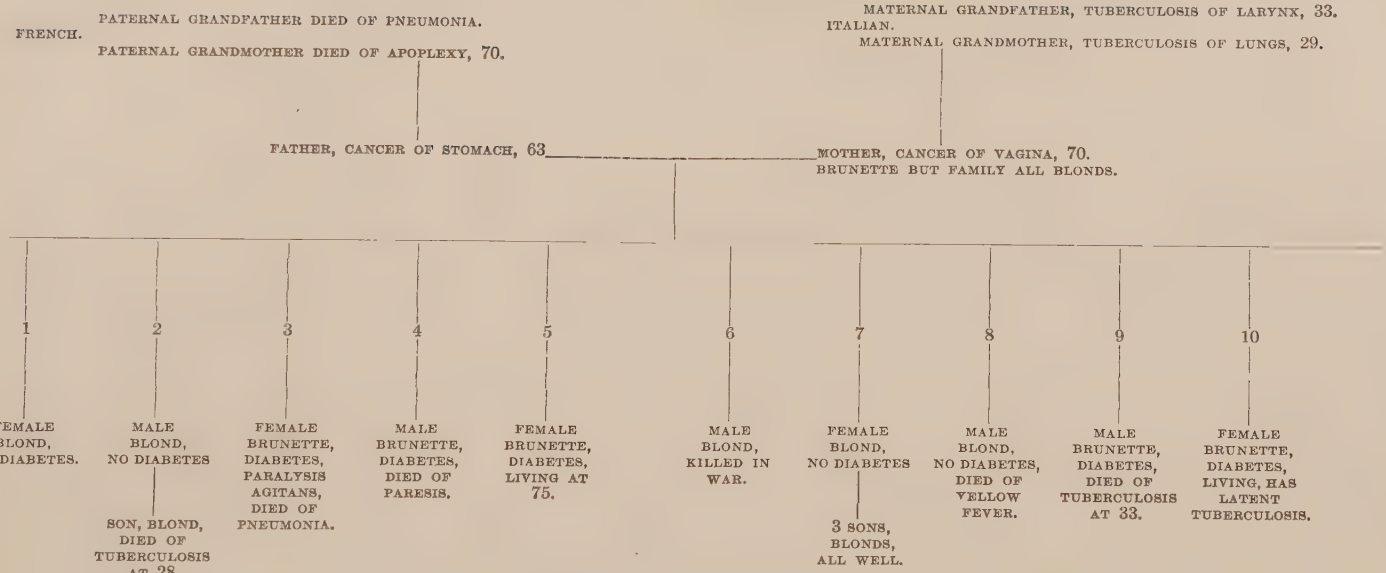
By H. R. M. LANDIS, M.D.
PHILADELPHIA

Facts relating to heredity in the transmission of disease are of sufficient interest at the present time to warrant placing on record the following observation:

Several years ago a woman, aged fifty-eight years, consulted me in regard to her lungs. In taking her history she stated that she had had glycosuria for a number of years. This, however, gave her little or no trouble provided she exercised reasonable care as to her diet. She furthermore volunteered the statement that several other members of her family also suffered from the same condition. The discovery of the glycosuria was accidental. The patient in question had applied, at the age of twenty-eight, for life insurance and was rejected because of the presence of sugar in the urine. At this time another sister was in ill health, and suspecting she might be suffering from a similar condition, her urine was examined and revealed the presence of sugar. This led to the remaining seven brothers and sisters submitting themselves to a similar test, with the rather remarkable result that the five brunettes were found to be diabetic while the four blonds (one blond having been killed several years before) were free from sugar.

An unusually satisfactory history was obtained as to the cause of death for four generations (see chart).

The following points are to be noted: (1) That of the nine members of the third generation, five were known to have a chronic glycosuria and that they were all brunettes. (2) That the remaining four were blonds and none of them showed the presence of sugar in the urine. (3) While both the father and mother were brunettes the other members of the mother's family were blonds. (4) The presence of tuberculosis in three of the four generations.


FRENCH.
PATERNAL GRANDFATHER DIED OF PNEUMONIA.
PATERNAL GRANDMOTHER DIED OF APOPLEXY, 70.
FATHER, CANCER OF STOMACH, 63
MATERNAL GRANDFATHER, TUBERCULOSIS OF LARYNX, 33.
ITALIAN.
MATERNAL GRANDMOTHER, TUBERCULOSIS OF LUNGS, 29.
MOTHER, CANCER OF VAGINA, 70.
BRUNETTE BUT FAMILY ALL BLONDS.
1
FEMALE BLOND, NO DIABETES.
2
MALE BLOND, NO DIABETES
SON, BLOND, DIED OF TUBERCULOSIS AT 28.
3
FEMALE BRUNETTE, DIABETES, PARALYSIS AGITANS, DIED OF PNEUMONIA.
4
MALE BRUNETTE, DIABETES, DIED OF PARESIS.
5
FEMALE BRUNETTE, DIABETES, LIVING AT 75.
6
MALE BLOND, KILLED IN WAR.
7
FEMALE BLOND, NO DIABETES
3 SONS, BLONDS, ALL WELL.
8
MALE BLOND, NO DIABETES, DIED OF YELLOW FEVER.
9
MALE BRUNETTE, DIABETES, DIED OF TUBERCULOSIS AT 33.
10
FEMALE BRUNETTE, DIABETES, LIVING, HAS LATENT TUBERCULOSIS.

DISCUSSION

Dr. S. Solis-Cohen, Philadelphia: This confirms in some measure certain speculations on the subject of tuberculosis and carcinoma on the one hand and diabetes and endocrine disturbances on the other, which I have frequently mentioned in discussion. In the *Osler Birthday Volume* I have recorded three clans, consisting of a number of families, traced in some instances to the fourth and fifth generations, in which we found distinct antagonism between diabetes and tuberculosis or carcinoma. Those who escaped tuberculosis and carcinoma usually showed marked endocrin and autonomic disturbances, some of them with polyuria, glycosuria or diabetes. Among a group of 65 diabetics, 1 showed tuberculosis. The proportion in the general community is given as 1 to 7, but even if it is only 1 to 20 the disproportion is marked. I am sure that analysis of large statistics will bear out the same fact—that relatively few tuberculous patients acquire diabetes and that relatively few diabetics acquire tuberculosis. In other words, tuberculosis and diabetes in descendants of a common stock are really manifestations of antagonistic tendencies; the natural reactions against tuberculosis and carcinoma being such as to excite, when in excess, autonomic and endocrine disorder, more or less marked, and of a character to predispose to errors of carbohydrate metabolism.

Dr. J. M. Anders, Philadelphia: I have been interested in the subject of familial diabetes. My rather extended experience in the treatment of obesity confirms the view that it occurs mostly in the obese. I have the definite impression that in the cases of familial diabetes which occur in corpulent subjects, gout figures as the cause of the obesity. The manifestations occur usually in late life and are not severe, but if, as they may rarely, in early life, they are severe. Van Noorden has shown a close connection between obesity and diabetes. Gout is in the same group. Obesity and gout are markedly hereditary, and in both there is a tendency to glycosuria. This partly, at least, explains familial diabetes.

Dr. F. M. Allen, New York: Studies of this kind furnish information on the question of how and why diabetic tendencies are inherited, whether they follow Mendelian or other known laws, etc., and they thus contribute toward solving the problem of diabetic heredity. Blood-sugar analyses and glucose tolerance tests are highly desirable also on the supposedly normal members of diabetic families for the recognition of the milder degrees of diabetic tendency. Studies of the pancreas of both the diabetic and the non-diabetic members at autopsy may also be important.

FURTHER OBSERVATIONS ON THE BLOOD-SUGAR TOLERANCE TEST AS AN AID IN THE DIAGNOSIS OF GASTROINTESTINAL CANCER

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AND
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In a study of the blood-sugar tolerance test as an aid in the diagnosis of gastrointestinal cancer presented by us last year the following conclusions were noted:

1. There is present in carcinoma of the gastrointestinal tract usually a rather characteristic curve of sugar tolerance which differs somewhat from that observed in carcinoma of other regions of the body. The curve of this affection usually presents a high sugar content even in the fasting state, followed by an initial rise up to 0.23 per cent or higher within 45 minutes after the ingestion of the dextrose, remaining at this level for at least 120 minutes, and rarely at any time during this period falling below 0. 2 per cent.

2. The sugar-tolerance test is rather distinctive, so that it may render valuable assistance as a means of differential diagnosis between carcinoma and other diseases of the gastrointestinal tract.

3. The opportunity has not been afforded to test a sufficient number of early cases of cancer of the stomach and intestines according to this method, so that as yet the value of this test as a means of early diagnosis has not been established; nevertheless, as positive curves occur equally whether cachexia exists or not, or whether the extent of the involvement be slight or great, we are under the impression that the results may be quite definite even in the early cases of the disease. This question, however, requires further study.

4. Finally, while we fully realize that this test is not specific of carcinoma and cannot be relied upon alone without entering into the clinical aspects of the disease, and that there are cases of carcinoma in which negative findings occur or non-malignant conditions in which the results are positive, nevertheless we are of the opinion that when properly performed the blood-sugar tolerance test may be of considerable diagnostic help in obscure cases of carcinoma of the gastrointestinal tract.

At that time our study included 32 cases of cancer of the gastrointestinal tract, all of which presented the typical cancer curve. This curve was, however, not observed in any one of 55 other examinations, including 5 cases of carcinoma outside of this tract, nor in 45 cases of benign affections of the gastrointestinal tract, including peptic ulcer, syphilis of the stomach, diarrhea, dysentery, achylia gastrica, chronic gastritis, cholelithiasis, chronic appendicitis, enteroptosis, nervous dyspepsia and mucous colitis.

The typical cancer curve can easily be distinguished from that observed in other affections of the gastrointestinal tract. It must be differentiated, according to our observations, from the normal curve, the atypical curve and the intermediate curve.

1. The Normal Curve. In normal individuals after the ingestion of 100 gm. of dextrose the blood-sugar content rises from 0.09 or even less to a height not usually above 0.165 per cent within 45 minutes, falling usually more gradually within 120 minutes to about the level as that observed in the fasting state. This character of curve was observed not only in normal individuals but also in the non-cancerous digestive disturbances other than those which have been classified in the following groups.

2. The Atypical Normal Curve. In certain individuals, often those affected with an achylia gastrica, an atypical normal curve is observed in which the blood-sugar content presents practically normal values in the fasting state, ascending to a height of from 0.16 to 0.18 per cent within 45 minutes after the ingestion of the dextrose, falling slowly within 120 minutes to a level of from 0.13 to 0.15 per cent, but not any lower.

3. The Intermediate Curve. The intermediate curve is usually observed in cases of carcinoma not associated with the

gastrointestinal tract, in which the blood-sugar content presents comparatively high values even in the fasting state of 0.14 to 0.17

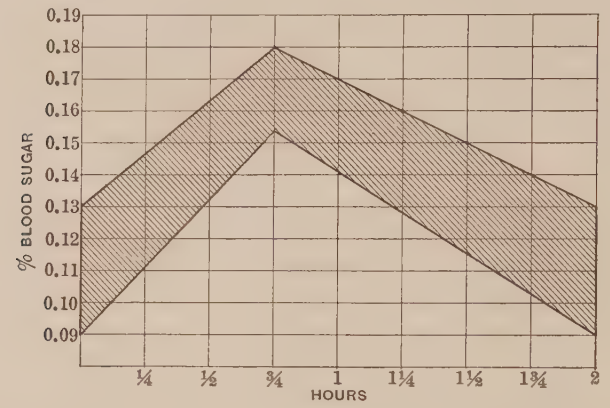

CHART I.—Maximum, minimum and average normal blood-sugar tolerance curve.

per cent, and in which after the ingestion of 100 gm. of dextrose there is an initial rise to a height of from 0.2 to 0.23 per cent within

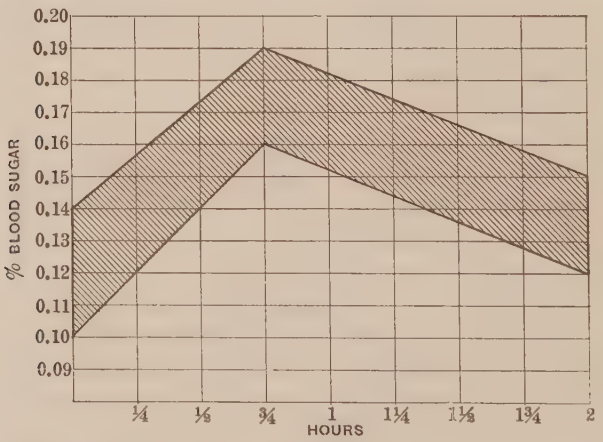

CHART II.—Maximum, minimum and average atypical normal curve.

45 minutes, followed within 120 minutes by a fall of from 0.18 to 0.21 per cent, never descending at this time to the level observed in the fasting state.

Since our former publication we have examined 43 additional cases of gastrointestinal cancer according to our method, totaling

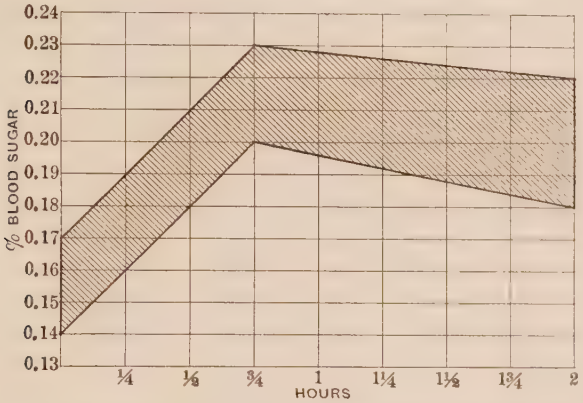

CHART III.—Maximum, minimum and average intermediate blood-sugar curve.

in all 75 cases. All were typical of the disease; of these 36 were operated on and the diagnosis was thus confirmed, and in the

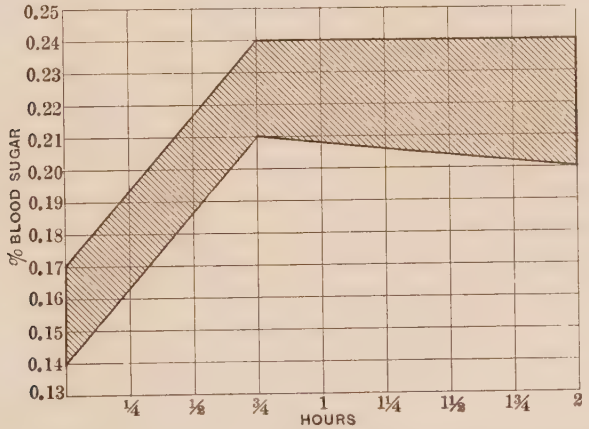

CHART IV.—Maximun, minimum and average blood-sugar tolerance curve in carcinoma of the gastro-intestinal tract.

remaining cases there were definite palpable abdominal masses in addition to the usual physical signs present in this affection, and the examination of the gastric contents as well as the roentgen-ray

findings and final outcome presented confirmatory evidence in every instance. Of these cases there were 47 of gastric carcinoma, of which there were 5 of cancer of the esophagus and cardia, 26 with obstructions at the pylorus, while 16 presented no obstruction. There were 5 cases of cancer of the colon, 2 of the pancreas, 8 of the liver, including 5 other abdominal organs; 3 of the rectum and 11 of carcinomatosis and 1 of the face and tongue.

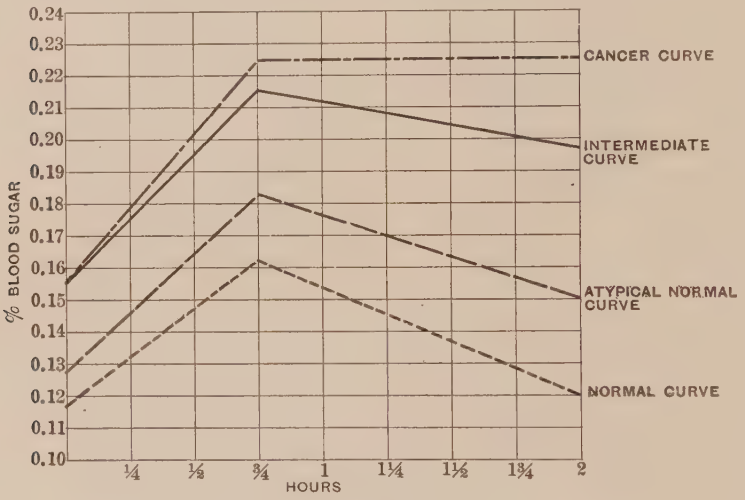

CHART V.

In our examinations we followed the plan described in our former communication with the patient on a fasting stomach. Blood was withdrawn for examination just before, in 45 minutes and 120 minutes after the administration of 100 gm. of dextrose and the blood sugar determined by means of the Epstein method, under the identical precautions described by us.

The character of curve present under these conditions in gastrointestinal cancer is quite characteristic and can usually be distinguished from that observed in cancer of any other region of the body or from that in non-malignant diseases. It was noted in all of these cases of cancer of the gastrointestinal tract but 3, of these 2 G. D. (No. 15) and M. P. M. (No. 50) were instances of general carcinomatosis and 1, C. I. (No. 33), a case of rectal carcinoma; in

all the diagnosis was confirmed by operation and the curves presented were typically normal.

On the other hand there were 5 cases proved by operation to be non-malignant, in which typical cancer curves were obtained; of these there was M. B. C., a case of tuberculous peritonitis; M. M. C., of gastric syphilis; B. H., of carcinomatosis; S. O., of cholelithiasis, and J. T. S., of gastric syphilis.

It is important to note that diabetes, nephritis, tuberculosis and disturbances of the thyroid should always be excluded before the tolerance test is undertaken, inasmuch as hyperglycemia is frequently present in these affections. However, as this test is to be utilized only as a means of differentiation between carcinoma and other diseases of the gastrointestinal tract, many conditions associated with hyperglycemia need not here be taken into consideration.

Inasmuch, therefore, as this test has proved positive in 72 of the 75 cases of cancer of the gastrointestinal tract, there can be but little question of its value as a diagnostic measure, even though it was found positive in 5 cases of non-malignant disease of these organs.

The value of this test is well illustrated in 3 cases of this series. We append a brief abstract of the histories of these cases:

Case 1.—S. B. (No. 2112), female, aged sixty-seven years, had been complaining of digestive disturbances for many years. Her appendix had been removed in 1908 with marked relief, but during the following year symptoms were again noted in the form of acute attacks of pain in the upper right quadrant, which were severe at times and were accompanied by nausea, vomiting, chills and fever, but without jaundice. Recently the attacks had become more frequent, but not quite so severe as formerly, although during the past two weeks there was continuous distress in the abdomen, accompanied with nausea and vomiting. Loss of flesh and great prostration was also noted. At this time the liver had become markedly enlarged and quite tender on pressure. The diagnosis of cholecystitis was made even though the blood-sugar tolerance test revealed a typical cancer curve. At operation numerous adhesions were detected in the upper right quadrant region, and the gall-bladder was found to contain a thickened, ropy infected bile, and cholecystectomy was performed. The patient recovered from the effects of the operation, but after several weeks nausea, vomiting and abdominal discomfort manifested themselves. Symptoms of pyloric obstruction were now noted with vomiting of the retention type. A month following the first operation a second was performed for the relief of the pyloric obstruction, when a large carcinoma was observed involving the pylorus and liver.

Case 2.—C. S. I. (No. 5091), male, aged fifty-four years, had been affected with indigestion for four months. At the onset he was taken with an acute attack of pain in the lower right quadrant of the abdomen and a diagnosis of acute appendicitis was made. Since then there had been constant indigestion, with general discomfort in the abdomen, pressure, distention, loss of appetite, eructations, extreme constipation and a loss of thirty pounds in weight. On palpation a mass was noted in the right lower quadrant with extreme tenderness in this area. The blood-sugar tolerance test revealed a definite cancer curve. At operation performed by Dr. J. M. T. Finney a carcinoma of the cecum was revealed and resection was performed; no metastases were noted. The patient made a good recovery, and when a subsequent blood-sugar tolerance test was made two months after operation a perfectly normal curve was revealed.

Case 3.—J. S. L. (No. 22677), female, aged sixty-six years, had been affected with abdominal pain and indigestion for some months, which had become aggravated during the past few weeks. There was pain in the right lower quadrant which was dull and dragging in nature, gradually becoming more and more intense; finally exquisite tenderness was noted in this region of the abdomen and a diagnosis of ovarian cyst with possible twisted pedicle was made. At operation on entering the abdominal wall an enormous pocket of pus was found from which a liter of pus exuded and a retrocecal abscess was discovered. A blood-sugar tolerance test made at this time revealed a typical cancer curve. The patient continued on with abdominal discomfort and slight temperature, notwithstanding the free drainage, and nausea, vomiting, progressive weakness and loss of flesh became prominent manifestations. The drainage continued, and when a larger incision was made carcinomatous infiltration of the intestine and peritoneum was observed.

TABLE PRESENTING THE PERCENTAGES OF BLOOD SUGAR OBTAINED IN THE 75 CASES OF CANCER OF THE GASTROINTESTINAL TRACT

1	A., F. C.	Cardia	.172	.260	.248
2	B., C.	Pylorus	.164	.224	.230
3	B., C.	Cardia	.192	.282	.250
4	B., C.	Stomach	.141	.193	.172
5	B., C.	Stomach	.140	.193	.172
6	B., I.	Pylorus	.121	.197	.178
7	B., B. B.	Pancreas	.159	.194	.198
8	B., M. J.	Pylorus	.141	.250	.238
9	B., G. F.	Liver	.120	.198	.192
10	B., S. S.	Stomach	.163	.268	.278
11	C., M. H.	Stomach	.196	.240	.203
12	C., M.	Stomach and liver	.146	.230	.242
13	C., T.	Stomach and liver	.147	.194	.191
14	C., G.	Stomach	.162	.204	.201
15	D., G.	Carcinomatosis	.142	.180	.150
16	D., B.	Carcinomatosis	.156	.260	.268
17	D., N.	Stomach	.170	.220	.224

18	D., J. A.	Carcinomatosis	.161	.258	.255
19	D., C. E.	Pylorus	.131	.187	.183
20	D., D. E.	Stomach	.178	.364	.332
21	F., S.	Rectum: Before operation	.198	.198	.200
		After operation	.148	.188	.184
22	F., M.	Liver	.120	.180	.200
23	F., T. W.	Pylorus	.154	.226	.272
24	G., M.	Carcinomatosis	.150	.186	.190
25	G., E.	Intestines and uterus	.125	.197	.194
26	G., D.	Carcinomatosis	.161	.250	.256
27	G., H.	Liver	.163	.214	.208
28	H., G.	Pylorus	.147	.192	.188
29	H., I. L.	Liver	.154	.210	.206
30	H., S.	Rectum and intestines	.172	.248	.280
31	H., W.	Pylorus	.149	.198	.194
32	I., G. S.	Cecum: Before operation	.155	.226	.258
		After operation	.082	.170	.110
33	I., G.	Rectum	.012	.167	.113
34	J., B.	Pylorus	.142	.187	.179
35	K., W. H.	Pylorus	.176	.282	.270
36	K., B.	Pylorus	.132	.198	.214
37	L., J.	Stomach	.148	.236	.232
38	L., J. E.	Stomach	.198	.276	.264
39	L., M. B.	Pylorus	.159	.191	.198
40	L., F. C.	Stomach	.170	.268	.208
41	L., F.	Rectum	.202	.300	.264
42	L., J.	Pylorus	.176	.252	.266
43	M., F.	Liver	.154	.200	.204
44	M., R. E.	Stomach	.142	.194	.192
45	M., P. J.	Pylorus	.116	.206	.192
46	M., K.	Prostate and intestines	.157	.198	.172
47	M., C. J.	Carcinomatosis	.158	.210	.210
48	M., M.	Pylorus	.138	.204	.204
49	M., H. T.	Pylorus	.136	.252	.280
50	M., M. P.	Carcinomatosis	.120	.168	.127
51	N., A.	Intestines	.154	.216	.220
52	P., G. G.	Stomach	.164	.242	.238
53	P., L.	Cardia	.168	.204	.192
54	P., M. G.	Stomach	.152	.198	.200
55	P., T.	Carcinomatosis	.171	.234	.242
56	P., H. G.	Stomach	.142	.262	.247
57	R., E.	Pylorus	.141	.192	.174
58	S., E. P.	Bladder and liver	.160	.245	.238
59	S., F. L.	Tongue	.384	.497	.497
60	S., C.	Carcinomatosis and liver	.162	.198	.200
61	S., I. P.	Carcinomatosis	.121	.162	.153
62	S., M.	Cardia	.131	.180	.224
63	S., C.	Stomach	.200	.266	.256
64	S., L.	Cardia	.178	.234	.228
65	T., J. T.	Pylorus	.171	.239	.240
66	T., M.	Carcinomatosis	.171	.214	.204
67	V., S.	Stomach	.138	.214	.206
68	W., C.	Pancreas	.160	.224	.198
69	W., C. C.	Pylorus	.156	.230	.218
70	W., L. H.	Stomach	.168	.290	.270
71	W., S.	Pylorus	.162	.198	.194
72	W., E.	Stomach	.154	.192	.178
73	W., V.	Pylorus	.164	.198	.224
74	Z., V.	Pylorus	.152	.236	.240
75	Z., W. A.	Pylorus	.161	.220	.218

Conclusion. From our further study of the blood-sugar tolerance test in malignant as well as in benign diseases of the gastrointestinal tract we are more fully convinced that this test may be utilized to great advantage as a means of differentiating between these affections, and that while it cannot in any way be considered specific for carcinoma, when taken into consideration with the other clinical evidence it may serve as a valuable aid in diagnosis in obscure cases of carcinoma of the gastrointestinal tract.

MANIFEST TETANY ASSOCIATED WITH AN ATTACK OF PAROXYSMAL HYPERPNEA IN A PATIENT CONVALESCENT FROM EPIDEMIC ENCEPHALITIS

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(ABSTRACT)

The patient, a young man of eighteen, was referred to us for diagnostic study on February 4, 1921. He complained of attacks of hard breathing, difficulty in sleeping and what he called slipping spells, in which he said he lost control for a few minutes and had but a faint recollection afterward of what happened during the spells. His parents who observed him during these spells said that he talked much during them and lost control of himself.

The mother, a very nervous woman, having lost three other children, has always been exceedingly solicitous regarding the patient's health. She sleeps in the same room with him at night, and becomes panicky if he shows the slightest symptom of illness. Despite this heredity and faulty environment the boy remained fairly well until January 24, 1920, when he was suddenly attacked with headache and chills. He felt better next day and engaged in a game of volley ball. During the game he became excited, accused another boy of cheating, became violent and had to be removed and carried home. He continued to be maniacal for the next fifteen days, and during this period had fever, the temperature averaging 102° F. He talked excessively day and night. There was some

diplopia. The fever and delirium gradually ceased but convalescence was slow. He complained of marked disturbances of digestion, and of insomnia and he suffered from peculiar paroxysms of deep breathing.

On account of the digestive disturbances he was operated upon in June, when a gangrenous appendix was found and removed. In September tonsillectomy was performed.

At intervals after his acute attack he manifested occasional mental lapses which he called "slipping spells." In these he feels drowsy and weak, though he remembers the attacks but feebly afterward. These attacks are probably petit-mal paroxysms.

During the taking of the anamnesis and the making of the physical examination the patient manifested recurring paroxysms of deep and labored respirations, each paroxysm lasting for two or three minutes.

The positive findings at the physical examination included slight anisocoria, the left pupil being larger than the right; slight double ptosis; slight nystagmus on strong lateral deflection of the eyes; slightly exaggerated deep reflexes. There were no other paralyses and there was no objective disturbance of sensibility. The eyegrounds were negative.

Toward the end of the examination another paroxysm of hyperpnea set in. The patient became excited, the respirations were very deep and the respiratory rate was accelerated. In a few minutes there was sweating of the face and fibrillary twitching of the left upper eyelid. Then, suddenly, the patient went into an acute attack of tetany with bilateral carpal spasm. During the attack he complained of numbness of the upper extremities. Chvostek's sign in the first degree could be elicited.

The attack persisted for from fifteen to twenty minutes, during which time a photograph was made.

The patient remained under observation in the nursing home for a few days, during which time *x*-ray examinations, routine laboratory tests and special neurological and psychiatric examinations were made. There was no return of the tetany during his stay, and even latent signs of tetany could not be demonstrated after the attack of manifest tetany had passed off.

The urine contained a trace of albumin. The blood, stomach juice and feces were normal. The cerebrospinal fluid contained three cells; the globulin test was negative as was also the Wassermann test.

X-ray examinations were negative except for a slight filling defect at the pylorus, suggestive of a small pyloric ulcer.

With rest, isolation, reassurance and dietetic measures the patient rapidly improved and returned to his home in North Carolina. Up to the time of this report there has been no return of the attacks of hyperpnea and no return of the tetany.

The interesting features of the case are (*a*) the psychotic-ophthalmoplegic type of epidemic encephalitis with epileptiform and psychoneurotic sequelæ, and (*b*) the paroxysms of hyperpnea with the occurrence of manifest tetany during one of the attacks.

The association of an attack of tetany with a paroxysm of pathologic hyperpnea interested us exceedingly, as we had just been reading the reports of Collip and Backus and of Grant and Goldman, who had observed symptoms of tetany during physiologic studies of voluntary forced respirations. Careful chemical control by these physiologists during the overventilation phenomena led them to the belief that the tetany was due to disturbance of the acid-base equilibrium. They found that during voluntary forced respirations the alveolar CO_2 tension fell, there was an alkalosis of the blood, a tendency to alkaline urine, a diminution in the ammonia output and a slight increase of the calcium content of the blood.

It will be recalled that in the earlier studies of tetany acidosis during the attacks was reported, whereas in some of the more recent studies of tetany, especially of experimental tetany (Wilson and Stearns; Wilson, Stearns and Janney), an alkalosis was observed. Certain other clinical observations of recent date are interesting in this connection. Howland and Marriott observed the occurrence of tetany in three infants after the administration of sodium bicarbonate, and Harrop observed tetany in an adult after the administration of sixty grams of sodium bicarbonate.

In the light of the physiologic studies referred to it would seem clear that the tetany in our patient during hyperpnea was a tetany of chemical origin dependent upon disturbance of the acid-base

equilibrium. The patient has no tetany otherwise. Even the latent signs of tetany were absent on the days following the attack.

Up to recently it has been supposed that all tetany is due to hypoparathyroidism. These newer observations of tetany associated with disturbances of metabolism, and especially of the acid-base equilibrium, make it seem probable that if there is a unitary pathogenesis for tetany this unity may lie in a metabolic vice that in some instances depends upon hypoparathyroidism and in other instances has a different etiology.

The case here reported in abstract will subsequently be published in detail.

THE ADRENALS AND THE THYROID IN TUBERCULOSIS

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For some time we have been giving attention to the role of the endocrine glands in experimental and clinical tuberculosis. During the past year, following an observation made in connection with a series of experiments [1 2] to determine the effect of removal of one adrenal upon the course of experimental tuberculosis, we have kept accurate records of the adrenal weights and body weights of many unselected tuberculous and non-tuberculous guinea-pigs. The major results are shown in the table and graph (Figs. 1 and 2).

FIG. 1.—ADRENAL WEIGHTS, ABSOLUTE AND RELATIVE TO BODY-WEIGHT

	Combined adrenal weight, grams.	Combined adrenal weight, per cent of body weight.	Increase from normal, per cent.
Normal	0.388	0.065	
Tuberculous	0.460	0.074	14
Septic infections and pneumonia	0.487	0.084	29
Tuberculosis less than six weeks	0.400	0.067	3
Tuberculosis more than six weeks (average ten weeks)	0.500	0.079	22

It is clear from these data that there is an absolute and relative increase of adrenal weight in the course of tuberculous disease and also in septic infections in these animals, and that in the case of tuberculosis it does not appear until the disease is well generalized,

roughly about two months after inoculation. Careful analysis of the figures has proved that this is not dependent on changes in body weight and that it is quite constant.

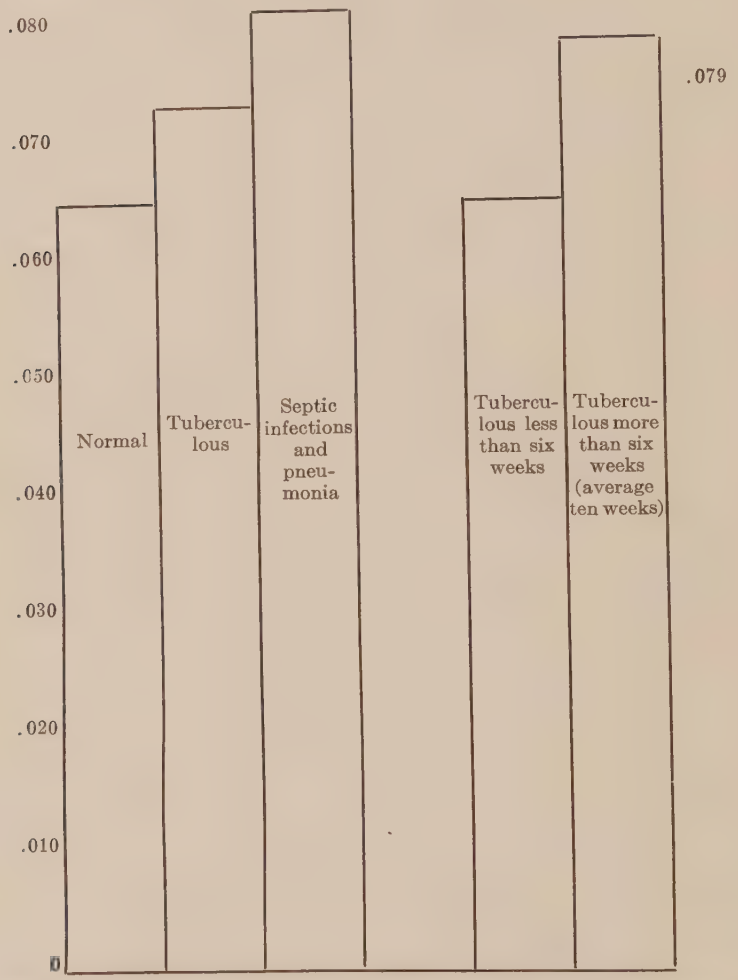

FIG. 2.—Combined adrenal weights expressed in percentage of body weight.

We have found a few closely related observations by other investigators. Calmette[3] reports enlargement and congestion of the adrenals following single fatal doses of tuberculin in guinea-pigs, and

states that he has observed sclerosis, which may eventually prove fatal, after repeated small doses. With Sézary and others he believes that a similar toxic atrophy and sclerosis, without the presence of tubercles or tubercle bacilli, may occur as a complication of pulmonary tuberculosis in man.

Elliott,[4] in connection with a study of the adrenalin content of the glands at autopsy in various diseases, observed that the adrenal weight was markedly increased in several cases of tuberculosis. The figures which he gives are from acute cases, but he states, also, that in chronic tuberculosis with general emaciation there is no corresponding exhaustion of the lipoids in the adrenal cortex. It is greatly to be hoped that he will continue his observations and report an extensive series of adrenal weights and body weights in human tuberculosis. He has found edema and increase of the glands to as much as double the normal size in fatal cases of acute pneumonia, and some of our highest weights have been in animals dying of an acute epizoötic pneumonia.

Microscopic sections in our guinea-pigs have shown tuberculous lesions of the medulla in one instance only. Apart from this there have been no clearly characteristic changes. The medulla does not appear to enlarge, and if there is true hyperplasia, rather than the mere cellular hypertrophy and increased vascularity of active function, it is probably cortical. When hypertrophy occurs in pregnancy, and following the removal of one adrenal, it is the cortex which enlarges (Stilling, Biedl, Crowe and Wislocki,[5] Elliott.[6]) There is an extensive and rapidly growing literature on the specific functions of cortex and medulla and their relative importance to life, but we have not time or space to review it here.

As to the direct clinical bearing of these observations on human tuberculosis there is as yet little evidence. We are inclined to interpret the adrenal enlargement as due to a demand for increased function. This is in accord with the views of Sergent[7] and Sajous[8] that the asthenia of tuberculosis is partly dependent on adrenal insufficiency and may be relieved by giving adrenalin, an observation which we have confirmed in our own practice. It seems probable, if it is the cortex which hypertrophies, that extract of adrenal cortex or of the whole gland may be still more beneficial, and we are trying these preparations now with some of our patients.

In the case of the thyroid we have more clinical observations but less accurate experimental data as yet. Many clinicians have reported changes in the thyroid in tuberculosis, the most constant of which is a slight hyperplasia with symptoms of hyperthyroidism in early cases, frequently followed by more or less atrophy in later stages (Turban,[9] Stanton,[10] Strouse,[11] Webb,[12] Sajous,[8] Coulaud[13 14].) The difficulty of distinguishing between early tuberculosis and hyperthyroidism, in many instances, is well known, and the similarity of symptoms has led to some confused thought and various contradictory explanations. It appears that these confusingly similar symptoms may be caused either by tuberculosis alone or by hyperthyroidism alone, and the Goetsch test, together with basal metabolism determinations, may be of real value in distinguishing between these two groups of cases, since uncomplicated tuberculosis apparently does not tend to give a positive test (Heise and Brown,[15] McBrayer.[16]) Yet the test may be positive in cases in which the real trouble is tuberculous but accompanied by this rather characteristic secondary hyperthyroidism.

We have noted enlargement of the thyroid in our tuberculous guinea-pigs while studying the effect of removal of increasing portions of the gland,[12 19] though as yet we have no figures ready to cite on this point. We lean to the same interpretation of this hypertrophy as in the case of the adrenals—namely, that it is a phase in the marshaling of the body's defences,[8] and that while it may reach a toxic degree its tendency on the whole is beneficial. Numerous clinical observations seem to support this view and have led us to believe that in general those patients whose thyroids show slight palpable enlargement do better than those with small inactive glands. This agrees with an observation of Plummer[17] at the Mayo Clinic that tuberculosis is much rarer in patients with exophthalmic goiter than in the general population. Kocher and after him C. H. Mayo[18] have noted a percentage and absolute increase of lymphocytes in hyperthyroidism, and we have confirmed this in a number of cases; and it is undoubtedly true, in our experience, that a high mononuclear index (the sum of the lymphocyte and endothelial leukocyte percentages in the differential blood count) is one of the most trustworthy signs of favorable prognosis which we have at our disposal in tuberculosis.

Here again we are interested in the therapeutic application. We feel that some cases of chronic tuberculosis have a real thyroid deficiency, that this interferes with their recovery and that it can be made good by the administration of thyroid extract. This must, of course, be undertaken with great caution, since there are patients to whom the drug is distinctly toxic.

REFERENCES

1. Webb, Gilbert, Hartwell and Ryder: Tr. Nat. Tuberc. Assn., April, 1920, p. 336.
2. Am. Rev. Tuberc., October, 1920, p. 605
3. L'Infection bacillaire et la tuberculose, Paris, 1920.
4. Quart. Jour. Med., Oxford, October, 1914.
5. Bull. Johns Hopkins Hosp., October, 1914, p. 287.
6. Practitioner, January, 1915, p. 123.
7. Etudes cliniques sur la tuberculose, 1908–1918, Paris, 1919.
8. New York Med. Jour., September, 1917, p. 389.
9. The Diagnosis of Tuberculosis of the Lungs (Trans. Morland), London, 1905.
10. Tr. Nat. Tuberc. Assn., 1905.
11. Med. Clin. of Chicago, January, 1917, p. 745.
12. Jour. Lab. and Clin. Med., March, 1916, p. 414.
13. Bull. Soc. méd. hôp., December 17, 1920, p. 1551.
14. Ibid., January 14, 1921, p. 5.
15. Am. Rev. Tuberc., October, 1920, p. 609.
16. Ibid., p. 616.
17. Personal communication to G. B. Webb.
18. Mayo Clinic Papers, 1910, p. 453.
19. Webb, Gilbert and Ryder: Am. Rev. Tuberc., May, 1921.

THE RELATION OF ACROMEGALY TO THYROID DISEASE, WITH A STATISTICAL STUDY

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(By Invitation)

The view that acromegaly is caused by hyperactivity of the anterior pituitary body after epiphyseal union has taken place is generally accepted. Cushing,[1] however, adduced experimental evidence to show that the disease is due to some perversion of secretion rather than merely to an excess of normal secretion.

With the slow and gradual development of the affection the sella turcica dilates into a cavity of variable size, as shown by an *x*-ray examination. In practically all cases of acromegaly that have come to autopsy, enlargement or disease of the hypophysis has been found, with a corresponding dilatation of the sella turcica and persistence of the thymus in most instances. The new growths of the pituitary gland may be of different character, *e. g.*, adenoma, sarcoma, carcinoma and cystic degeneration. It is generally held that the majority are adenomata. Sternberg[2] states that the acute form of acromegaly is due to a rapidly progressive tumor, frequently sarcoma, of the hypophysis. The enlargement may be due to functional hypertrophy with or without hyperplasia of the chromophile cells.

On the other hand it has been shown that destructive lesions of this gland may be at times unaccompanied by clinical symptoms of acromegaly.[3] In 1891 one of us (Anders) and Henry W. Cattell

reported to the Philadelphia Neurological Society a hemorrhagic tumor of the pituitary body and infundibulum which had been unattended with any signs of acromegaly during life.

Conversely, Arnold and Ponfick[4] have reported cases of acromegaly with the pituitary glands normal in all respects. It is true, as pointed out by Friedman,[5] that all ductless-gland disturbances may manifest themselves clinically without gross or microscopic changes. Hemmeter[6] has also emphasized the occurrence of endocrine syndromes without actual pathologic alterations. These are termed "ductless-gland neuroses." Among predisposing factors in acromegaly, the role of some of which are still in doubt, are race, age, sex, heredity and trauma, to which detailed reference will be made hereafter.

In 1886 Rogowitsch,[7] and later Stieda[8] and Hofmeister,[9] noted hypertrophy of the pituitary after removal of the thyroid in rabbits. Bourneville and Bricon[10] state that in sporadic cretinism in which the thyroid is lost the pituitary has been found to be enlarged. Boyce and Beadles[11] and other observers found an increase in size of the pituitary gland in hyperthyroidism, while Abram[12] noted changes in this gland in a case of carcinoma of the thyroid. Marie and Marinesco[13] described a case of acromegaly in which there was simple hypertrophy of the thyroid and thyroid-like tissue of the hypophysis. Schöneman[14] in 85 cases of goiter found marked changes in the pituitary in all but one instance.

Conversely in animals subjected to experimental hypophysectomy, definite histologic alterations in the thyroid have been observed. For example, Caselli[15] found hypertrophy of the entire thyroid apparatus. The view of Comte[16] and Hoskins[17] that these two glands can function vicariously for one another is supported by the results of total or nearly total extirpation of the hypophysis and those of thyroidectomy.

Of 15 cases of pituitary disease in Cushing's series, in which the thyroid was examined microscopically, all showed "a low epithelium with excess of colloid." It should be noted that at the time of his examinations all of the cases were evidencing signs of glandular insufficiency.

Exner[18] relates that in two of Hochenegg's cases of acromegaly,

after partial extirpation of the pituitary, enlargement of the thyroid occurred. Says Cushing,[19] "It is my impression that the gland is most liable to show enlargement in individuals with clinical evidences of past hyperpituitarism"—suggesting, in other words, that the same underlying biochemic factor causes a hyperplasia of both structures rather than that the thyroid assumes a compensatory and vicarious role for the hypophysis. Observations upon tumors of the hypophysis were described as early as 1810, when Wenzel[20] tried to link epilepsy with diseases of the pituitary. To Friedreich[21] we owe the first attempt to formulate the symptoms of pituitary new growths.

Cushing[22] has described cases of acromegaly in which with overgrowth and overactivity of the anterior lobe are associated symptoms of posterior lobe insufficiency, *i. e.*, adiposis, increased sugar tolerance, polyuria, polydipsia, subnormal temperature, dry skin, loss of hair, epileptiform disturbances and the like.

Pineles[23] has emphasized the clinical relationship between acromegaly and myxedema, as shown, he believes, by the comparatively frequent coexistence of the two conditions, by the similarity of many of the symptoms and by the occurrence of similar pathologic changes in the two conditions. Without doubt these two glands exhibit points of resemblance both from a histologic and a physiologic viewpoint, so that their commonly observed clinical association should be expected. To show the similarity of their effects upon metabolism it is only necessary to call attention to the fact that myxedema occurring before the growth of the bones is completed causes an arrest of ossification. Functional associations between the pituitary, adrenal, thymus, pineal and genital glands are also recognized, but these will not receive consideration here for want of time and space.

Whether in the cases of associated changes in the pituitary and thyroid the latter shows atrophy or hypertrophy in the majority of the cases is, perhaps, no longer a moot question (*vide infra*). For example, Schönemann[24] believes that an increase of hypophysis may take place in cases of hypertrophy of the thyroid gland; he has, however, "Failed to find any compensatory increase of the hypophysis in goiter—that is, in cases of degeneration of the

thyroid or in cachexia strumipriva." On the other hand, Souza-Leite,[25] based on an extensive collection of cases, holds that an atrophy of the thyroid is more frequently associated with an enlarged hypophysis than hypertrophy of the thyroid. Hutchinson[26] affirms that the thyroid is atrophied in about 10 per cent of all cases of acromegaly and enlarged in 30 per cent, the enlargement being due usually to cystic degeneration.

It would appear that practically all of the larger earlier post-mortem reports of fatal myxedema in which the hypophysis was examined no abnormalities were observed. We venture to believe, however, that solitary instances of the association of lesions under discussion is decidedly more common than was formerly supposed. Moderate increase in size and microscopic structural changes which are not infrequently present are easily overlooked unless the examinations be thorough and complete.

The effects of hyperactivity of the anterior hypophysis differs in different cases with alterations in the functions of the other internal glands, more particularly the thyroid gland. In this connection the observation of Berkeley[27] that the anterior lobe of the hypophysis is abundantly supplied with sympathetic fibers while the posterior lobe is devoid of such fibers, goes far toward explaining the interrelation between diseases of the former and hyper- and hypothyroidism.

In a previous paper by us[28] the purpose was, among other things, to collect all cases of pituitary disease recorded in literature covering a period of five years terminating December 31, 1913, and to aid in clearing the question of the relation of hypo- and hyperthyroidism to acromegaly on the one hand and glycosuria on the other. In 5 out of 16 cases (31.5 per cent) of acromegaly in which the thyroid was reported no changes were noted. Of these the thyroid was small in 1, normal in 1, sclerotic and cystic in 1, not palpable in 1, while the remaining case (our own) showed well-marked evidences of hyperthyroidism. These figures would appear to show that the role played by hyperthyroidism in cases of acromegaly is trivial and uncertain. On the other hand they indicate that hypothyroidism is more commonly found in association with pituitary disease.

It is of much interest to note that while acromegaly is regarded as being a rare disease six years after Marie[29] first described the condition, Collins[30] was able to collect about 90 cases from the literature. In 1898 Hinsdale[31] found reports of 130 cases, of which 23 showed exophthalmos, and in the same year Woods Hutchinson collected 218 cases.

In 1893 James J. Putman[32] treated a case of acromegaly with thyroid powder (gr. xv every second day); the condition improved markedly, the enlargement of the hands and feet having practically disappeared. We believe it is safe to infer that in this instance hypothyroidism was associated and that the hypertrophy affected principally the soft tissues. Putman refers briefly to two additional cases, "The one stands nearer to acromegaly, the other to myxedema, though in both the diagnosis is doubtful." Both were given thyroids with excellent results. In the former there was a large swelling of the neck, "Probably a thyroid, which is diminishing under thyroid extract." There can be but little doubt that this was an instance of combined pituitary and thyroid disease, since hypothyroidism is known to occur in subjects showing enlargement of the gland. Such cases we believe are common examples of advancing hypopituitarism associated with hypothyroidism. They are, as a rule, greatly benefited by careful glandular therapy.

On the other hand Burns's, Benson's and Mosse's cases (quoted by Hinsdale) treated with thyroid extract showed improvement principally in the subjective symptoms, more particularly in the nervous phenomena, without diminution in the size of either the lips, hands or feet. In such an absolute diagnosis of an associated hypothyroidism would be unwarranted. In this connection it is to be borne in mind that in myxedema the hypertrophy affects the soft tissues exclusively, although the basal metabolism, especially as related to the nervous system, is decreased in this disease. Improvement in the mental condition, however, in the absence of a favorable effect upon the soft tissues in myxedema is contrary to universal experience

The growing tendency to regard individual cases of glandular disease as being of pluriglandular origin is unjustified. As pointed out by Falta[33] in speaking of pathologic correlation, "The disease

that has first involved one gland involves in its further course other members of the ductless glandular system." True it is that upon close analysis of the history, symptoms and course of a given case it is frequently possible to discern which ductless gland was first affected.

Secondary to primary involvement of the hypophysis the symptomatic indications of both hypo- and hyperactivity of the thyroid may arise. That the converse may obtain, however, seems to be supported by ample evidence, *e. g.*, in certain cases of Graves's disease. Bell[34] has studied the pituitary in disease of the thyroid. He described the results of a histologic examination in a case of cretinism in a female who died at the age of thirty-three years; the structural characteristics found indicated considerable activity, which he thinks were probably compensatory in character.

In exophthalmic goiter many more basophils than is normal are found in the pars anterior, while in the pars posterior the number is normal. Certain it is, however, that reliable deductions must await further information concerning secondary pluriglandular effects. At all events it would seem to us highly probable that the effect upon the pituitary (anterior lobe and intermedia) is more marked after the removal of the thyroid than after the removal of other hormonopoietic organs.

Friedman[35] claims there is a certain degree of hypophyseal hyperactivity in hyperthyroid and of hypoactivity of the pituitary in hypothyroid states. If this be true, then, pituitary products are contraindicated in the treatment of Graves's disease, but indicated in addition to thyroid in myxedematous subjects. This dictum receives confirmation from the histologic studies of the hypophysis in hyperthyroidism and myxedema by Fry,[36] who points out that in the former condition the histology corresponds to a gland of high activity; in the latter to a gland of low activity. The pituitary in myxedema is anatomically a hypertrophied gland, but physiologically an atrophied one.

It is especially in these hybrids that mild forms of hypothyroidism, which are so difficult to diagnose, are liable to occur; but their recognition is important, as shown by the happy effects of metabolic stimulation by means of thyroid extract so far as the

myxedematous manifestations are concerned. As elsewhere stated by one of us:[37] "In both the skin is dry; the subcutaneous fibrous connective tissue is thickened by overgrowth; the tongue is enlarged; the mucous membranes, especially of the nose, soft palate and uvula, are thickened; and menstrual disorders in the female; in both there is irritability of temper, slowness of the mental processes and thick, difficult speech."

Certain obvious features, however, belong to each of these diseases alone when found to be combined. For example, the general bony enlargement with separation of the condyles, spreading apart of the teeth, increased growth of the hair on the head, enlarged joints, the severe headaches and increased muscular strength followed by muscular weakness; in women hairiness assuming the male type, disturbances of vision and bilateral hemianopsia are dependent on the acromegaly; while the tendency to chilling, a subnormal temperature, diminished metabolism, elastic skin—dry and scaly (no pittings)—miniature thyroid, malar flushes, loss of hair, slow small pulse, often arteriosclerosis and slight albuminuria, as well as a peculiarly distorted physiognomy (masked facies), belong to myxedema. Attention should be directed here to those minor forms in which the symptoms are merely suggestive of associated hypothyroidism. In such the cautious use of thyroxin is advised in order to confirm the diagnosis.

In associated hyperthyroidism the cardinal symptoms, such as tachycardia, tremor, exophthalmos, nervousness, increased sweating and increased metabolism, may be more or less frankly expressed, in which case an assured diagnosis is beyond question. In certain cases the degree of hyperthyroidism is moderate or even slight and the symptoms presented are indefinite and variable. Slight tachycardia, a moist delicate skin, a tendency to diarrhea, often polyuria, amenorrhea, slight increase in size of the thyroid, and somewhat widened palpebral fissures and an increased metabolic rate, which is a reliable criterion, are among the most significant diagnostic features in these mild, atypical forms. In the sympathicotonic type one may observe exophthalmos with tachycardia, but no sweats or diarrhea, marked falling out of the hair or alimentary glycosuria.

In this connection Janney's[38] view that the classic symptoms of exophthalmic goiter are of toxic origin or due to a perverted rather than to mere excess of thyroid secretion is to be recollected. If we accept this theory we shall be in a position to appreciate how toxic and hypothyroid symptoms may be combined in a given case. Again, in cases of acromegaly with associated exophthalmic goiter and deficiency symptoms small doses of thyroid extract may prove useful while larger ones would produce an unfavorable effect. Before forming a final diagnostic opinion, however, exophthalmos due to the pressure effects of the hypophyseal enlargement must be eliminated. It is to be recollected that a state of thyroid insufficiency following hypophyseal overactivity is not reached before death in all cases, but only in those in which degenerative changes occur.

An examination of the literature reveals not a few cases of combined pituitary and thyroid disease in which the manifestations of hypothyroidism overshadow those of hypophyseal disease. To overlook the true nature of these is unfortunate in view of the happy effects of appropriate glandular therapy. We have personally observed modifications of the function of the thyroid in two cases of acromegaly, which are here briefly detailed:

Case I.—A. J. M., female, aged thirty-four years; single; weight, 168 pounds; teacher in the public schools. Referred to one of us for study and treatment by Dr. S. Lewis Ziegler November 3, 1917. The paternal grandfather died of apoplexy; also the maternal grandfather. The patient had some of the childish diseases and during adolescence repeated attacks of tonsillitis. Eight years ago she was operated upon for tuberculosis of the "right tube and ovary;" the wound is still unhealed (fecal fistula). The patient had been taking little exercise and menstruation had always been delayed and scanty, followed by amenorrhea during the last fourteen years. For a long time she had been constipated and complained of nervousness, irritability and annoyed by any noise; during the past year she had much headache and always felt chilly. During the past two years she noticed divergence of the left eye and states that for about eight years she had large bones.

Status Praesens. Bones of the face, hands and feet, as well as those of the trunk, unusually large; hands and feet especially broad in the bony parts, spade-like; lower jaw prognathous. Complains of severe headaches

at times with nervousness and marked irritability of temper; speech is slow, thick and leathery.

Myxedematous infiltration of the skin of the face and extremities is present, but had not been previously recognized. An associated symptomatic anemia is more marked than usual. For example a blood count showed: Erythrocytes, 3,110,000; leukocytes, 6000; color index, 9; hemoglobin, 55 per cent. The skin exhibits a slightly yellowish hue with marked pallor of the mucous membranes. The thorax is negative; a fecal fistula is noted in the right lower quadrant of the abdomen. The thyroid is not palpable. The urine proved negative and there is increased sugar tolerance.

Dr. Ziegler reports eye-findings as follows: "She first noticed diplopia about one year ago, which doubtless was caused by the muscular error in her eyes, which amounts to a hyperphoria of 4 degrees and an exophoria varying from P 18 degrees to P 20 degrees. Her refraction is compound hypermetropic astigmatism of low grade. I have corrected the hyperphoria but cannot correct the exophoria without again causing her diplopia, as one eye wanders off far enough to limit her to monocular vision. The nerve head shows slight pallor on the temporal side. The fields are absolutely hemianopic with some contraction, as is indicated in the charts of the same, which I enclose."

An *x*-ray study by Dr. G. E. Pfahler follows: "I find great thickening of the skull. The cranial vault is approximately 2 cm. in thickness—three or four times as thick as normal. There is a tumor in the region of the pituitary and this tumor has extended both upward above the sella and has broken through the floor of the sella, extending into the region of the sphenoid. This is the first time that I have seen such a condition (in which the tumor has extended through the floor). The clinoid processes are not widely separated or destroyed, as is commonly the case, probably due to the fact that the tumor has extended downward more than upward. The hands show the thickening about the ends of the distal phalanges, and in fact general thickening about the ends of all of the phalanges and of all of the metacarpals. They have the appearance that we commonly find in acromegaly." The case is looked upon as one of acromegaly with associated myxedema and tuberculosis.

The use of thyroid extract in small doses has been remarkably effective for good. Not only has there been a marked subsidence of all of the myxedematous features, but the headaches, speech and nervous irritability have also been greatly relieved. The bony skeleton, however, has remained stationary.

Case II.—A. H., aged forty-six years; female; occupation, cook; weight, 170 pounds; height, 5 feet 9½ inches. First came under the care of our surgical colleague, Dr. William L. Rodman, in the Medico-Chirurgical

Hospital, December 5, 1913, at which time she was suffering from a gangrenous wound, inflicted by a fish-bone, of the left hand. A few days later the patient was referred from the medical service to us for study and treatment. Polydipsia, polyuria and glycosuria, as well as bulimia, were all prominent features throughout the illness. The percentage of sugar at first was 5 per cent, but at the end of one week it rose gradually to 7 per cent, and remained at this level until the end.

The typical symptoms of acromegaly were in evidence. According to the history the bones of the hands and face began to enlarge four years ago while the glycosuria set in about two years later. An *x*-ray examination by Dr. Pfahler showed marked lipping at the edges of the articular surfaces of the metacarpal bones and phalanges, such as is found in acromegaly, and also a tumor of the pituitary gland, with absorption of the clinoid processes. The diagnosis of associated hyperthyroidism was quite apparent from the noticeable enlargement of the thyroid gland, together with tremor, tachycardia and exophthalmos. The indications of exophthalmic goiter were most probably developed after the glycosuria had existed for an indefinite period of time, although being, as is usual, of insidious origin it was impossible to fix the precise date of onset. The Wassermann reaction gave a negative result.

With a view of indicating the true pathologic relationship between disease of the hypophysis and thyroid the results of a statistical study are subjoined. The total number of cases collected by us, which was 215, includes only a few that were reported prior to the publication of the paper of Dr. Hinsdale in 1898 and that of Hutchinson in 1902. Table I is a detailed tabulation of 91 cases in which thyroid disturbance was associated.

It should be pointed out that the notations to be found in the columns headed hyper- and hypothyroidism are the inferences drawn by the original reporters, with few exceptions. Failure to record the sugar tolerance and condition of the thyroid gland and its function in numerous cases is to be regretted. It is interesting to note that all of our series or 215 cases occurred in the white race with a single exception, which was reported by Yamada, of Tokio (*vide* Table I). Trauma played a role in 3 cases and in 2 the disease followed "psychic trauma." According to these findings the influence of traumatism as a disposing factor is insignificant. There can be no doubt but that a family type exists, although the influence of heredity must, on the whole, be trivial, as evidenced by our results, namely, two instances only of the entire series.

TABLE I

Reporter.	Reference.	Age.	Sex.	Lesions in thyroid.	Hyperthyroidism.	Hypothyroidism.	Lesions in pituitary.	Glycosuria.	Remarks.
Lancereaux	Semaine méd., 1895, xv, 61	40	F	Hypertrophy	Present	....	....	Present	Trauma.
Henrot	Quoted by Lancereaux, loc. cit.	36	M	Goiter	Present	....	Tumor size of hen's egg	Present	Reported in 1877; heart hypertrophied.
Valat	Idem.	..	..	Goiter	Present	....	....	.....	Reported in 1893.
Haskovec	Wien. klin. Rundschau, 1895, ix, 257	35	M	Atrophy	....	....	....	Absent	Head injury followed by meningitis.
Sears	Boston Med. and Surg. Jour., 1896, cxxxv, 16	45	F	....	....	Present	....	Absent	Improved by thyroid administration.
Dodgson	British Med. Jour.,1896, i, 660	23	F	Enlarged					
Worcester	Boston Med. and Surg. Jour., 1896, cxxxiv,413	70	F	Not palpable	....	Marked	Sarcoma with psammomatous degeneration	.....	Myxedema disappeared under thyroid treatment.
Furnival	Lancet, London, 1897, ii, 1190	58	M	Slightly enlarged and altered in function	....	....	Adenoma-cystic degeneration		
Dalton	Idem.	..	..	Normal, except thyroid juice unusually fluid	....	Present	Sarcoma or hypertrophy and posterior lobe almost disappeared	Present	Thymus normal.
Murray	Edinburgh Med. Jour., 1897, i, 170	37	F	Goiter	Present	....	....	Present	Pulmonary tuberculosis.
Murray	Idem.	63	F	Large cystic goiter	Absent	....	....	Absent	
Murray	Idem.	34	F	Enlarged	Present	....	Enlarged	Present	Pulmonary tuberculosis; thymus hypertrophied.
Schiff	Wien. klin. Wchnschr., 1897, x, 277	37	M	....	....	Present	....	Increased tolerance	

Schiff	Ztschr. f. klin. Med., 1897, xxxii, 289, Suppl.	..	..	....	....	Present	....	....	Unable to consult article
Osborne	Tr. Assn. Am. Phys., 1897, xii, 262	47	M	Enlarged 101 gm., extrathyroid in chest, 36½ gm.	....	....	Sarcoma (?)	.....	Heart enormous.
Comte	Beitr. z. path. Anat. u. z. allg. Path., 1898, xxiii, 90	46	F	Much enlarged	Present	....	Enlarged 0.415 gm.		
Comte	Idem.	32	F	Enlarged 390 gm.	Present	Absent	Enlarged 0.85 gm.		
Bailey	Philadelphia Med. Jour., 1898, i, 789	65	F	Parenchymatous hypertrophy; nearly 3 times normal weight	....	....	Adenoma		
Hunter	Tr. Path. Soc., London, 1898, xlix, 246	52	M	Hypertrophy	....	....	Hypertrophy		
Neal	Ibid., p. 237	41	F	Parenchymatous goiter 6½ oz.	Absent	....	Hypertrophy	.....	Slight improvement under thyroid.
Coe	Jour. Am. Med. Assn., 1898, xxxi, 1347	68	F	Small	....	....	....	Absent	Homicidal.
Thompson	Med. Press and Circ., London, 1898, n. s., lxv, 377	19	F	....	....	Present	....	Absent	
Ponfick	Ztschr. f. klin. Med., 1899, vol. i.	47	M	Marked atrophy	....	Present	Chronic interstitial inflammation and atrophy		
Lediard	British Med. Jour., 1903, i, 789	43	F	Bilateral goiter (large)	....	....	....	Previous glycosuria	Left lobe removed with improvement in pressure symptoms.
Cattle	Ibid., p. 780	30	F	Goiter	Present	....	....	Absent	Exophthalmos, tachycardia and excessive sweating.
Snell	Ibid., ii, 131	38	F	Not enlarged	....	Present	....	Absent	Improved under thyroid treatment.
Launois and Roy	Nouv. iconog. de la Salpêtrière, Paris, 1903, xvi, 163	36	M	Hypertrophy; 250 gm.	....	....	Large epithelioma	Present	

TABLE I.—(*Continued.*)

Reporter.	Reference.	Age.	Sex.	Lesions in thyroid.	Hyperthyroidism.	Hypothyroidism.	Lesions in pituitary.	Glycosuria.	Remarks.
Heising	Wisconsin Med. Jour., 1903–04, ii, 684	38	F	Small	....	....	....	.....	Preceded by severe psychic trauma; thymus persistent.
Bate	Louisville Month. Jour. Med. and Surg., 1903–04, x, 374	28	F	....	Probable		....	.....	Attributes tachycardia (100) to overaction of thyroid-like tissue in pituitary.
Salomon	Berl. klin. Wchnschr., 1904, xli, 635	49	M	....	Probable	....	....	.....	Increased metabolism; skin resembles myxedema.
Salomon	Idem.	42	M	....	Present	....	....	.....	Increased metabolism; skin resembles myxedema; marked arteriosclerosis.
Greene	New York Med. Jour., October, 1905, p. 846	25	M	Small	....	Present	....	Absent	Myxedema disappeared under treatment.
King	South. California Pract., 1905, xx, 410	37	M	....	Absent	Previous myxedema	....	Absent	
Kerry	Ophth. Rev., London, 1905, xxiv, 193	50	M	....	....	Probable	....	.....	Marked improvement under thyroid treatment.
Ballet and Laignel Lavastine	Nouv. iconog. de la Salpêtrière, Paris, 1905, xviii, 176	70	F	Hyperplasia	....	....	Parenchymatous hypertrophy	.....	Hyperplasia of adrenals
Taylor	W. London Med. Jour., 1905, x, 124	34	F	....	....	Probable	....	.....	Sent as case of myxedema.
Schoenborn	Beitr. z. path. Anat. u. z. allg. Path., 1905, Suppl., vii, 468	65	M	Cystic goiter with interstitial inflammation	....	....	Adenoma size of walnut	Absent	Cardiorenal asthma.
Porter	Ophth. Rec., Chicago, 1906, xv, 267	47	F	Greatly enlarged	Present				

Curren	Maritime Med. News, Halifax, 1907, xix, 98	20	F	"Thickening in region of thyroid"	Present				
Haywood	Homeopathic Eye Jour., 1907, xiii, 48	33	F	....	Present	. . .	....	. . .	Improved by pituitary treatment; atrophy of ovaries and uterus.
Sainton and Rathery	Soc. méd. d. hôp., Paris, 1908, xxv, 647	32	F	Atrophy and sclerosis	....	Present	Malignant tumor	Absent	Thymus persistent.
Renon and Delille	Ibid., p. 973	47	F	....	....	Present	Tumor	Absent	
Kocher	Deutsch. Ztschr. f. Chir., Leipsig, 1909, c, 13	30	F	Small	....	....	Sarcoma	Present	Thymus persistent; operation.
Carmody	Lancet, London, 1909, i, 1599	33	F	Previous goiter	....	....	....	Absent	Skin thick but could be pinched, now abnormally active; no improvement under pituitary treatment.
Exner	Mitt. a. d. Grenzgeb. d. Med. u. Chir., 1909, xx, 620	30	F	Enlarged after operation on pituitary	....	....	Malignant adenoma	Tolerance increased	Tongue, lips, hands and feet smaller after operation.
Exner	Idem.	34	F	Isthmus became size of walnut after operation	....	....	Malignant adenoma	Tolerance increased	
Packard	Am. Med., 1910, n. s., v, 539	36	F	Hypertrophy	....	....	*X*-ray unable to demonstrate tumor		
Grove	Bull. Johns Hopkins Hosp., 1910, xxi, 290	47	F	Colloid goiter	....	Present	Enlarged	Absent	Partial thyroidectomy.
East	Jour. Mental Sci., 1912, lviii, 631	43	F	Enlarged	....	....	....	.....	Mental symptoms.
Cushing	Pituitary Body and its Disorders, Philadelphia, 1912, p. 30	35	M	Small	Absent	Absent	Adenoma	.....	Acromegaly secondary to gigantism; exophthalmos due to neighborhood pressure.
Cushing	Ibid., p. 36	26	F	Colloid goiter	Absent	Absent	Adenoma		
Cushing	Ibid., p. 85	36	M	Large and colloidal	Absent	Absent	Tumor	.....	Slight acromegaly followed by hypopituitarism; trauma.

TABLE I.—(*Continued.*)

Reporter.	Reference.	Age.	Sex.	Lesions in thyroid.	Hyperthyroidism.	Hypothyroidism.	Lesions in pituitary.	Glycosuria.	Remarks.
Cushing	Ibid., p. 137	40	M	Slightly enlarged	Absent	Absent	Slight enlargement of sella turcica	.....	Hypertrichosis.
Cushing	Ibid., p. 140	38	M	Colloid goiter	Absent	Absent	Chromophil-struma		
Cushing	Ibid., p. 150	59	M	"Plunging goiter"	Former	Absent	Enlarged	.....	Goiter removed.
Cushing	Ibid., p. 154	42	M	"Possibly enlarged"	Absent	Probable	....	.....	Benefited by thyroid treatment; complicated by gigantism, hay fever and tuberculous adenitis.
Cushing	Ibid., p. 194	33	M	Enlarged; excess of colloid	Absent	Absent	"Functional hyperplastic change"	.....	Secondary to hydrocephalus.
Holmes	Proc. Roy. Soc. Med., London, 1912–13, vol. vi, Neurol. Ophth. Sect., X	36	F	....	....	Probable	Sella turcica enlarged	.....	Improved under thyroid treatment.
Garrod	Ibid., lxxviii	..	..	....	....	Present	Sella turcica enlarged	.....	Improved under thyroid.
Brown	Ibid., lxxxvii	40	F	Cystic goiter	....	"Possible myxedema"	....	Tolerance increased	Previous acromegaly, now hypopituitarism; some improvement under thyroid.
Bartlett	Tr. Chicago Path. Soc., 1913, ix, 56	25	M	Colloid goiter	....	....	Chromophil-cell adenoma	.....	Thymus persistent.
Gordinier and Kirk	Albany Med. Ann., 1913, xxxiv, 189	39	F	Distinctly enlarged	....	....	Tumor		
Grinker	Jour. Am. Med. Assn., 1913, lxi, 235	49	M	Absent	..	Present	Large adenoma	Intermittent	Followed infected vaccination and psychic trauma; benefited by thyroid; epilepsy.

Millioni	Riforma med., Naples, 1913, Nos. 39, 40 and 41, vol. xxix	34	F	Small	....	Present	Marked enlargement; probably tumor	Absent	
Fernsides	Proc. Roy. Soc. Med., London, 1913–14, vii; Electro-therap. Sect., 46	40	F	Not palpable	....	Present			
Dunn	Am. Jour. Med. Sci., 1914, cxlviii, 214	61	M	Small	....	....	....	Tolerance increased	Hemoptysis; non-tuberculous; angioneurotic edema.
Anders and Jameson	Ibid., p. 323	46	F	Goiter	Present	....	Tumor	Present	Gangrene.
Osborne	Proc. Connecticut State Med. Soc., 1914, p. 164	40	M	....	....	Present			
Chizet	Lyon méd., 1914, cxxii, 882	15½	M	....	....	Present	Tumor	.....	Polyuria.
Bendell	Albany Med. Ann., 1915, xxxvi, 441	45	F	Colloid enlargement					
Fürth	Med. klin. Berl., 1915, xi, 1347	40	F	Palpation negative	....	Deficient iodine production	Tumor	.....	Temporary improvement under iodide treatment, father had goiter.
Falta	Ductless Glandular Diseases, Philadelphia, 1916, p. 237	32	M	Enlarged	....	....	....	Tolerance increased	Heart enlarged; hypertrichosis.
Falta	Ibid., p. 243	31	M	Slightly enlarged	....	....	....	Tolerance increased	Hypertrichosis.
Falta	Ibid., p. 243	31	M	Not enlarged	Probable	....	Adenoma	.....	Hypertrichosis; thyroid in aggravated symptoms; temporary improvement after operation.
Falta	Ibid., p. 246	33	F	Enlarged	....	....	....	Tolerance decreased	Marked hypertrichosis; lues.
Falta	Ibid., p. 253	37	F	....	Present	....	Grayish-red tumor	Tolerance decreased	
Falta	Ibid., p. 266	31	F	....	Present	Present	....	Tolerance decreased	Hyper- followed by hypothyroidism.

TABLE I.—(*Continued.*)

Reporter.	Reference.	Age.	Sex.	Lesions in thyroid.	Hyperthyroidism.	Hypothyroidism.	Lesions in pituitary.	Glycosuria.	Remarks.
Futcher	Med. Clin. North America, 1917, i, 131	67	F	....	Absent	Possible association of myxedema	Sella turcica enlarged	Tolerance increased	Possible early paralysis agitans.
Soner	Med. Jour. So. Africa, 1918, xiii, 102	34	F	....	Absent	Present	....	.....	Improved under thyroid treatment.
Yamada	Mitt. a. d. Med. Fac. d. k. Univ. z. Tokyo, 1918, viii, 411	17	M	Hypertrophy	....	....	....	.....	Thymus persistent; died of beriberi.
Good and Ellis	Endocrinology, 1918, ii, 431	58	M	Atrophy	....	Present	Hemangio-endothelioma	Tolerance increased	Thymus persistent; testes and adrenals atrophied and sclerotic.
Lewis	Laryngoscope, 1918, xxviii, 604	34	F	....	....	Present			
Loewenberg	Internat. Clinics, 1918, 28th ser., iii, 154	34	M	Slightly enlarged	....	....	Tumor (calcified)	.....	Pulmonary tuberculosis.
Roth	Berl. klin. Wchnschr., 1918, lv, 305	28	M	Enlarged	....	....	Tumor	Absent	Developed copious milk secretion.
Howard	Am. Jour. Med. Sc., 1919, clviii, 830	31	M	Slight cystic degeneration	....	Probable	Round-cell sarcoma	Tolerance increased	
Howard	Idem.	47	F	Slight goiter	Probable	....	....	Present	
Webster	Arch. Radiol. and Electro., London, 1919–20, xxiv, 261	..	..	Enlarged	....	....	....	Tolerance increased	Headaches relieved by *x*-ray.
Tierney	Med. Clin. North America, 1920, No. 3, iv, 799	40	M	Enlarged	....	....	....	Tolerance increased	Father and sister acromegalic types.
Barr	New York Med. Jour., 1920, cxi, 248	36	M	Enlarged	....	....	Sarcoma	.....	Imbecile.
Bernstein	Not published	39	M	Small	....	Probable	Large adenoma	Tolerance increased	Operation relieved headache, preserved vision; acromegaly stationary since.
Anders and Jameson	Not previously published	34	F	Small	....	Present	Tumor	Tolerance increased	Improved by thyroid treatment.

TABLE II.—TOTAL CASES OF ACROMEGALY, 215

Age (cases mentioned, 195).		Sex (cases mentioned, 194).		Lesions of pituitary (cases mentioned, 99).	Lesions of thyroid (cases mentioned, 68).	Glycosuria (cases mentioned, 91).	
Male.	Female.	Males.	Females.			Present.	Absent.
37.14	38.76	90 (46.4 per cent)	104 (53.6 per cent)	"Tumor" . . . 41 Adenoma . . . 15 Sarcoma . . . . 12 Sella turcica enlarged 13 Enlarged . . . 6 Hypertrophy . . 3 Various 9	Enlarged . . . . 24 Goiter . . . . 20 Atrophy . . . . 17 Hypertrophy . . 7 Functional . . . 23	32 (35.2 per cent)	59 (64.8 per cent)
Average age, 38.3							

TABLE III.—COMBINED ACROMEGALY AND THYROID DISTURBANCE, 91 CASES

Age (cases mentioned, 86).		Sex (cases mentioned, 86).		Lesions of pituitary (cases mentioned, 47).	Lesions of thyroid (cases mentioned, 68).	Glycosuria (cases mentioned, 47).	
Male.	Female.	Males.	Females.			Present.	Absent.
39.64	39.98	37 (43 per cent)	49 (57 per cent)	"Tumor" . . . 13 Adenoma . . . 11 Sarcoma . . . . 6 Enlarged . . . . 4 Sella turcica enlarged 4 Hypertrophy . . 3 Various 6	Enlarged . . . . 24 Goiter . . . . 20 Atrophy . . . . 17 Hypertrophy . . 7 Functional . . . 23	14 (30 per cent)	33 (70 per cent)
Average age, 39.85							

TABLE IV.—ACROMEGALY WITHOUT THYROID DISTURBANCE, 124 CASES

Age (cases mentioned, 109).		Sex (cases mentioned, 108).		Lesions of pituitary (cases mentioned, 52).	Glycosuria (cases mentioned, 44).	
Male.	Female.	Males.	Females.		Present.	Absent.
35.4	37.67	53 (49 per cent)	55 (51 per cent)	"Tumor" . . . . . 28 Sella turcica enlarged . . . 9 Sarcoma . . . . . 6 Adenoma . . . . . 4 Cyst . . . . . 2 Enlarged . . . . . 2 Degeneration 1	18 (41 per cent)	26 (59 per cent)
Average age, 37.18						

A review of Table II shows that in 99 cases out of the 215 cases of acromegaly the lesions of the pituitary are mentioned and that in 68 cases thyroid changes are recorded. It is noteworthy that only three more examples of adenomata than sarcomata are reported among the pituitary lesions, it being currently stated by the older as well as recent writers that the great majority of the new growths of this gland in acromegaly are adenomata. The influence of sex upon the incidence of the disease is shown by a slight preponderance in favor of the female. On the other hand Hinsdale's figures indicate that the disease is more common in the male than in the female sex; for example, he found 73 males and 57 females in his series of 130 cases. The average age here given (thirty-eight and three-tenth years), which represents that of the ages reached at the time the cases were reported, is too high, but the date of onset was so frequently indeterminable from the histories recorded in the literature as to make this course necessary. Table III needs no account.

Table IV shows the age incidence to be slightly higher in the combined pituitary and thyroid cases than in the uncombined acromegalics and also a slightly greater preponderance in the female as compared with the male sex by comparison with acromegaly without associated thyroid lesions. It cannot be stated, however, that these figures afford a basis for the claim often advanced that the thyroid is a true sex gland, as is shown by both Graves's disease and myxedema when they occur independently of the acromegaly. For example, both the latter conditions are six to eight times as frequent in the female as in the male. It will be noted that the proportion of cases of adenoma in Table IV as compared with sarcoma is almost three times greater than in Table III, although the figures are too small on which to base any inferences.

TABLE V.—AGE BY DECADES

Acromegaly without thyroid disturbance		Acromegaly with thyroid disturbance	
Under 20 years	3	Under 20 years	3
21 to 30 "	32	21 to 30 "	7
31 to 40 "	34	31 to 40 "	39
41 to 50 "	17	41 to 50 "	25
51 to 60 "	18	51 to 60 "	4
61 to 70 "	4	61 to 70 "	6
71 to 80 "	1	71 to 80 "	2

An inspection of Table V, which presents the age incidence by decades, will show that for acromegaly with associated thyroid disturbance this is considerably higher than in the cases without involvement of the thyroid—an interesting fact. The explanation is probably to be found in the accepted view that thyroid complaints, myxedema in particular, develop later in life than uncomplicated acromegaly, as a rule. This table indicates clearly that the great majority of cases of acromegaly with thyroid disturbance arise between the thirtieth and fiftieth years of age, and that both during the decade prior to that period of life and subsequently the cases are far less numerous than in acromegaly without thyroid disturbance. Again, it should be noted that in the cases of acromegaly in which thyroid disturbance is combined the latter usually arises a number of years after the onset of the former, *i. e.*, when the stage of hypopituitarism is reached.

TABLE VI.—THYROID FUNCTION, 52 CASES

Hyperthyroidism 20 (38.5 per cent)		Hypothyroidism 32 (61.5 per cent)	
Male	5	Male	12
Female	14	Female	18
Glycosuria		Glycosuria	
Present	7	Present	2
Absent or increased tolerance	1	Absent or increased tolerance	16

Table VI exhibits the facts bearing on the relative incidence of hyper- and hypothyroidism and on the carbohydrate tolerance as well as the influence of sex on the thyroid function. It will be seen that in the hyperthyroid group the ratio in favor of the female is about 3 to 1 while in the hypothyroid cases it is 2 to 2. Again, the incidence of hypothyroidism as compared with that of hyperthyroidism is about as 3 to 2. With regard to the question of carbohydrate tolerance in cases of combined acromegaly and disturbed thyroid function Table VI demonstrates that in hyperthyroidism the sugar tolerance was decreased in 7 out of 8 cases—ratio 7 to 1. On the other hand in the cases of associated hypothyroidism sugar was absent or tolerance increased in the ratio of 8 to 1. It should be added that of the 16 cases belonging to the latter group in 7 the increased sugar tolerance was determined by the usual tests.

SUMMARY. Our literary studies have revealed an unexpected frequency of associated disturbance of the thyroid function in cases of acromegaly, or 42.8 per cent, as shown by Table I. It is highly probable that in many instances of pituitary disease with its usual syndrome the coëxistence of thyroid alterations and symptoms resulting therefrom were overlooked, as was true of one of the two cases, which was reported by us. Our investigations indicate that hypothyroidism is more commonly associated with acromegaly than hyperthyroidism, and that those combined cases which manifest myxedematous features are decidedly improved as the result of the use of thyroid preparations.

It follows that the recognition of the indications of hypothyroidism in connection with acromegaly is a matter of the utmost importance. In cases of acromegaly in which merely suspicious features of either hypo- or hyperthyroidism exist it is strongly urged that the metabolic rate be determined. The approved sugar tolerance test should be carried out in all cases of acromegaly with a view of determining the state of both the pituitary and thyroid function. Nothing of special significance bearing upon the etiologic factors of the disease under consideration resulted from our studies, with the possible exception of the facts developed in connection with the age groups by decades.

BIBLIOGRAPHY

1. The Pituitary Body and its Disorders, p. 10.
2. Nothnagel's Handbook, 1897, p. 7.
3. Jokoloff: Virchow's Archives, vol. xiii.
4. Quoted by Cushing: Loc. cit., p. 20.
5. New York Med. Jour., 1921, cxiii, 370.
6. Ibid., 1914, xcix, 101.
7. Modern Medicine (Osler and McCrae), 1915, iv, 803.
8. Ueber das Verhalten der Hypophysis des Kaninchens nach Entfernung der Schildrüse, Ziegler's Beiträge, vol. x.
9. Fortschr. d. Med., 1892, No. 4.
10. Arch. d. Neurol., 1886.
11. Bacteriol. and Pathol., 1892, i, 223 and 359; February, 1893. Pathologic Reports, University College, London, 1892–93, vol. i.
12. Liverpool Med.-Chir. Jour., July, 1896.
13. Anatomie pathologique de l'Acromegalie, Arch. d. méd. exp., 1891.
14. Virchow's Archives, 1892.
15. Riv. sper di fren., 1900, p. 468.
16. Beitr. z. path. Anat. u. z. allg. Path., 1898, xxiii, 90.
17. Am. Jour. Med. Sci., 1911, cxli, 374, 535.
18. Beiträge zur Pathologie und Pathogenese der Akromegalie, Mitt. a. d. Grenzgeb. d. Med. u. Chir., 1909, xx, 620–644.
19. Loc. cit., pp. 280–281.

20. Ueber den Hirnanhang fallsüchtiger Personen, Mainz, 1810.
21. Beiträge zur Lehre von den Geschwülsten innerhalb der Schädelhöhle, Wurzburg, 1853.
22. Am. Jour. Med. Sci., March, 1913, p. 313.
23. Volkmann's klin. Vort., 1899, p. 242.
24. Quoted by Boyce and Beadles: Loc. cit.
25. L'Acromegalie, Paris, 1890.
26. New York Med. Jour., 1898, No. 11, vol. lxvii.
27. Quoted by Bell, p. 32.
28. Am. Jour. Med. Sci., 1914, cxlvii, 323.
29. Revue de Méd., 1886.
30. Jour. Nerv. and Ment. Dis., New York, 1892.
31. Acromegaly, 1898, p. 23.
32. Tr. Assn. Am. Phys., 1893, viii, 338.
33. The Ductless Glandular Diseases, second edition, p. 440.
34. The Pituitary, William Wood & Co., New York, p. 262.
35. Loc. cit.
36. Quart. Jour. Med., 1914–1915, iii, 277.
37. Anders: Diagnosis of Myxedema, Am. Jour. Med. Sci., 1920, clx, 801.
38. Arch. Int. Med., August, 1918, p. 187.
39. This case is more fully reported in a previous article by Anders and Jameson, loc. cit.

MULTIPLE POLYPI IN THE STOMACH (GASTRIC POLYPOSIS)

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The following case came under observation recently: A woman, aged about fifty, no children. She has never been robust and has had many anxieties connected with outside affairs as well as with her household duties. For years her appetite was indifferent and she had daily discomfort in the epigastrium after meals. The bowels seldom acted without laxative drugs. As a result of all these causes she was weakly and nervous. Sleep was light and easily disturbed. She was thin and worn-looking. The abdomen was flat and rather rigid. There were tender areas in various parts, shifting from time to time, not persistent in any part, not even in the epigastrium, in which nothing abnormal was to be felt. The various reflexes were overactive.

There was nothing abnormal to be found connected with the lungs and heart. The urine was rather scanty and concentrated, but she drank little water. Gastric contents an hour after some food consisted of a little almost clear mucus, quite anacid. No food or blood was present.

A period of rest under capable nursing care appeared essential to her improvement, but before entering upon such a course it was decided to have an *x*-ray examination made of the gastrointestinal tract to make sure there were no signs of ulcer or chronic appendicitis. Observation immediately after the barium meal showed a remarkable scolloping along the greater curvature throughout its entire length (Fig. 1). Six hours later about one-third of the meal was still in the stomach. In addition to the irregular greater curva-

ture there were two or three oval areas appearing through the barium and on moderate pressure other areas were seen, doubtless due to the polypi on the anterior and posterior surfaces of the stomach coming into contact, permitting the rays to penetrate and become apparent on the screen (Fig. 2). The polypi evidently existed over all parts of the mucous membrane from the cardiac orifice nearly to

FIG. 1

the pylorus, but they did not obstruct the latter. Their presence throughout the whole stomach precluded the possibility of relief by surgical means.

She was placed under good nursing care, with as liberal a diet as she could take, and given hydrochloric acid and strychnin freely, with but little improvement after two months of such care.

The prognosis is of course infavorable but depends much on her care and the degree in which her nutrition can be maintained.

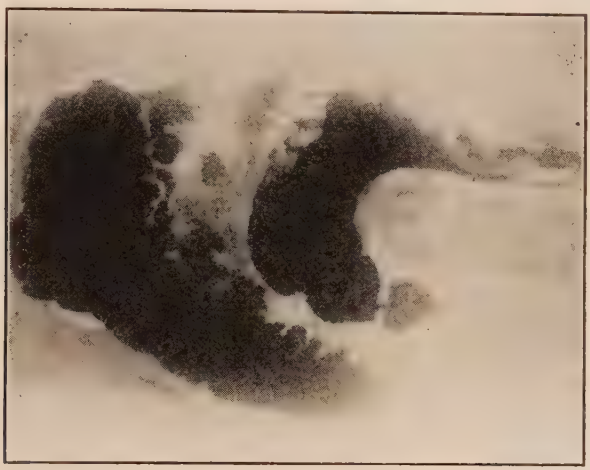

FIG. 2

The rarity of the disease is indicated by the fact that only two cases are on record in America. The first was reported in the *Journal of the American Medical Association*, 1913, vol, lxi, by J. A. Myers. In this case a mass of polypi obstructed the pylorus. The mass was

FIG. 3.—From Ewing's neoplastic diseases, p. 631.

removed, but unfortunately the hemorrhage could not be completely arrested, and death occurred three days later. The second

one was seen by D. C. Balfour,[1] of the Mayo Clinic, the first occurring in their large clinic. Also by Carmen and A. Miller.[2]

The polypi were disposed in irregular rows and varying in size from 0.5 to 2 cm. Their surfaces were smooth, containing no glands. Their pedicles were thick, made up of greatly hypertrophied mucous membrane, muscularis mucosa and a loose connective-tissue core from the submucosa, and contained fairly large arteries and veins. The depressions between the polypi consisted of mucous membrane in deep folds (Fig. 3). There was an abundant secretion of mucus, antacid and without digestive property.

There is nothing in clinical history or physical examination of such cases on which to establish a diagnosis—all the symptoms occur in various organic and functional conditions. In advanced cases there may be hæmatemesis and a sense of resistance over the stomach, especially the pylorus, the part most commonly affected. If lavage is done and tissue particles or polypi found they would be an indication of the condition. Fortunately, however, an *x*-ray examination gives us the aid necessary to permit of an early and certain diagnosis being made.

I have pleasure in acknowledging the kind assistance of Dr. W. H. Dickson, chief radiologist for gastroenteric diseases in the Toronto General Hospital, who made the first and second illustrations, and offered valuable advice in the diagnosis.

[1] Surg., Gynec. and Obst., 1919, xxxvii, 465.
[2] Roentgen Diagnosis of Diseases of the Alimentary Canal, 1920.

THE BLOOD IN TETRACHLORETHANE POISONING*

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Tetrachlorethane is a substance that readily poisons man, causing a fatal toxic hepatitis when it enters the body in sufficient amounts. It is used commercially because of its peculiar physical and chemical properties. It is a solvent for cellulose acetate, and is both waterproof and non-inflammable. Tetrachlorethane has risen to a point of paramount interest in the last few years because of its use in some of the so-called aëroplane "dopes," the varnish with which the linen wings of aëroplanes are painted.

During the war numerous cases of poisoning from use of this substance were reported among those engaged in the manufacture of aëroplanes in England, America and Germany. Some deaths occurred from this substance in England and Germany, but none have been reported from America. Tetrachlorethane is being utilized in this country in the manufacture of such articles as non-inflammable films, various lacquered goods and artificial silk.

The president of an artificial silk plant, employing about sixty persons at one time, requested one of us to supervise for five months an investigation of the health of his employees, aiming particularly to learn how to prevent any serious form of tetrachlorethane poisoning. The general clinical aspects of this investigation have

* From the Medical Laboratories of the Massachusetts General Hospital. This is paper No. 2 of a series of studies on the physiology and pathology of the blood from the Harvard Medical School and allied hospitals, a part of the expense of which has been defrayed from a grant from the Proctor Fund, of the Harvard Medical School, for the Study of Chronic Diseases.

been conducted by Dr. Derric C. Parmenter, whose report will be found in the April, 1921, issue of the *Journal of Industrial Hygiene*. The part of this general investigation that is reported here was undertaken by us to determine whether tetrachlorethane produced any abnormality of the blood, and if so, whether mild poisoning could be detected or even anticipated by blood examination. It was hoped that a blood examination might be of value in aiding the regulation of employment in such a plant.

Because tetrachlorethane may produce a toxic hepatitis somewhat similar to that which may be produced by arsenobenzol, salvarsan, etc., it was thought that the blood of tetrachlorethane workers might show increases of large mononuclear cells, as Evans[1] and one of us has observed occasionally occurs with the initial stages of poisoning from these arsenic products. It was also thought that blood changes in the nature of destruction of red cells with increased marrow activity might occur like those due to trinitrotoluene,[2] dinitrophenol and dinitrobenzene, substances which produce a toxic hepatits.

Before discussing the blood of the sixty-eight individuals whom we have studied, who were exposed and some of whom were poisoned by tetrachlorethane, it is desirable to briefly summarize the symptomatology and pathology of tetrachlorethane poisoning.

From the literature, which is chiefly made up of reports by Willcox[3] and others[4 5 6 7] (England), a summary by Hamilton[8] (U. S. A.), and by reports from Jungfer,[9] Grimm, Heffter and

[1] Observations on the Origin and Status of the So-called "Transitional" White Blood Cell, Arch. Int. Med., 1916, xvii, 1.

[2] Minot, G. R.: Blood Examinations of Trinitrotoluene Workers, Jour. Indus. Hygiene, 1919, i, 301.

[3] Willcox, W. H.: A Fatal Case of Poisoning by Tetrachloride of Ethane, Lancet, 1914, ii, 1489. An Outbreak of Toxic Jaundice due to Tetrachlorethane Poisoning; a New Type among Aëroplane Workers, Lancet, 1915, i, 544. The Treatment of Toxic Jaundice due to Tetrachlorethane Poisoning, British Med. Jour., 1916, i, 300.

[4] Legge, T. M.: Origin, Symptoms, Pathology, Treatment and Prophylaxis of Toxic Jaundice Observed in Munition Workers, Royal Soc. Med. Rep. (Sections of Medicine, Pathology and Epidemiology), January, 1917, p. 2.

[5] Spilsbury, B. H.: Origin, Symptoms, Pathology, Treatment and Prophylaxis of Toxic Jaundice Observed in Munition Workers, Royal Soc. Med. Rep. (Sections of Medicine, Pathology and Epidemiology), January, 1917, p. 42.

[6] Lee, W. E.: A Fatal Case of "Dope Poisoning," Lancet, 1916, i, 24.

[7] Barlow, F.: Dope Poisoning, Med. Press and Circ., 1916, cli, 471.

[8] Industrial Poisoning in Aircraft Manufacture, Jour. Am. Med. Assn., 1917, lxix, 2037.

[9] Tetrachlorethanvergiftungen in Flugzeugfabriken, Zentralbl. f. Gewerbehyg., 1914, ii, 222.

Joachimoglu,[10] and Koelsch[11] (Germany) we obtain a fairly good clinical picture of particularly the severer tetrachlorethane poisoning.

The individuals whom we have studied have given us a clearer picture of the early symptomatology of this form of industrial poison than obtained from the literature.

The earlier symptoms consist of three chief types—general, gastric and nervous. A typical case begins with a sense of abnormal fatigue; he perspires freely; drowsiness develops with loss of appetite, nausea, vomiting, constipation and headache. After a matter of days, or even weeks, jaundice develops and the gastric and nervous symptoms increase. There then may be confusion, delirium, coma and death. The preliminary symptoms of abnormal fatigue, general discontent and nervousness, inability to concentrate, loss of appetite, headache and insomnia constitute a clearly defined picture of the beginning of poisoning. These symptoms may develop over a period of several days or over a period of a few weeks. The insidious onset of such rather minor symptoms is a distinct feature, and they serve as a warning of the severer symptoms which may follow unless the individual is removed from exposure to the poison. Owing to their insidious onset and mild character these minor symptoms are readily overlooked.

Severer symptoms consist of an increase of constipation, nausea and vomiting, and in addition gas in the stomach, generalized abdominal pain, dizziness and very slight jaundice. Such disturbances may appear suddenly, but this is unusual. Still more advanced symptoms occurring with increase of jaundice are those of cholemia, which occur associated with severe necrosis of the liver cells, very similar to those of acute yellow atrophy. No other important pathological change has been reported.

Besides jaundice, which may become moderate in degree, the important physical signs which develop relatively late consist chiefly of an occasional palpable and tender liver, general abdominal tenderness, pallor and loss of weight.

[10] Gewerbliche Vergiftungen in Flugzeugfabriken, Vrtljschr. f. gerichtl. Med., 1914, Series 3, vol. xlviii, second supplement, pp. 162 and 191.

[11] Gewerbliche Vergiftungen durch Zelluloidlacke in der Flugzeugindustrie, München. med. Wchnschr., 1915, lxii, 1567.

Immediately following removal from exposure the symptoms do not necessarily subside but not infrequently increase for a few days. If the symptoms are of the very mildest degree they may disappear within two or three days, even without removal from exposure. Jaundice, even though not severe, when it is once established may persist for several weeks or even months before disappearing.

There were no severe cases of tetrachlorethane poisoning among the 68 employees of the artificial silk plant whom we studied during a period of five months. Twenty of these individuals were women and were practically not exposed to the poison. The others were men exposed to a greater or lesser degree at one time or another. Among them occurred 12 who showed symptoms best interpreted as very mild preliminary symptoms of poisoning, which persisted for but a few days. Their symptoms consisted of an abnormal sense of fatigue, irritability constipation and headache. There were nine others who showed more definite symptoms of poisoning. Two of these were moderately sick, but not to a degree to be considered serious or alarming, and not at any time sick enough to warrant their going to bed.

In all, 275 critical Wright-stained blood-smear examinations of the white cells, red cells and platelets were made on 68 employees: 60 white cell counts (37 persons) and 44 red cell counts (30 persons) and Tallquist hemoglobin determinations were made.* A few other observations on the blood were made but yielded no significant information.

The only reference that we have found in the literature to the blood in tetrachlorethane poisoning is by Willcox,[3] who merely states that "the jaundice is hepatogenous, not hemolytic, and that no appreciable anemia or changes in the red blood corpuscles occur."

The blood of certain employees whom we have studied showed distinct abnormalities, the most important alteration from normal being an increase of the large mononuclear cells.†

* It is a pleasure to acknowledge the technical assistance given by Miss Margaret Weld.

† In this paper, large mononuclear cells (Mallory's endothelial leukocyte) refer to those cells classically twice as large as a polynuclear neutrophile, which are often considered to originate in the bone-marrow, and from endothelium, to which the inappropriate term of transitional cell has been applied, especially to the more

Nearly all of the 25 individuals whose blood showed this change to a definite degree had or soon developed, especially if there was a progressive increase of these cells, some symptoms attributable to poisoning. On the other hand none of the 21 employees who had symptoms due to tetrachlorethane failed to show increases of the large mononuclear cells, which often formed over 30 per cent of the white corpuscles. There are other less marked and less significant changes that we have found in the bloods of individuals poisoned by this chemical.

Table I consists of an analysis of a typical case of poisoning. It shows the progression of symptoms eventuating in jaundice which remained for several weeks after the other symptoms had disappeared. The table also shows that as the symptoms increase progressive alterations in the blood develop, which returns toward normal as the symptoms subside. The blood changes particularly consist of the following five features: (1) The progressive increase of the large mononuclears. (2) The progressive increase of young large mononuclears, some formed and some broken. (3) The somewhat elevated white count. (4) The slight but progressive anemia. (5) The slight increase of platelets.

All of the 68 individuals studied have been divided into three groups, as shown in Table II, dependent on the one hand upon the number of large mononuclear cells in the blood and on the other upon the symptoms that occur. In this table the figures and data shown represent an average obtained at the time that the large mononuclear counts were the highest in each case.

Group I consists of all employees, 25 in number, who showed an increase of large mononuclear cells, above 12 per cent in a differential count of not less than 200 white cells. Twelve per cent has been chosen rather arbitrarily, but chiefly because of the fact that 10 per cent of large mononuclears may occur in the absence of any

mature form with the indented nucleus. These cells are very similar to those large mononuclear cells that probably originate in lymphatic tissue; but they are usually larger, more sharply defined, more granular and stain more distinctly than the large mononuclear cells of lymphatic origin. It is possible that, owing to the liver pathology of tetrachlorethane poisoning, the large mononuclear cells in these poisoned individuals have come from the endothelium of the liver as well as from the marrow. Both Dr. C. H. Bunting and Dr. J. H. Wright have been kind enough to examine some of the preparations and agree that the cells are of the type referred to.

clearly recognized pathology, and this number of these cells occurred in several people in the other groups. All of the 25 persons in Group I manifested clinical symptoms of poisoning at one time or another except 4. These 4 individuals have been placed in a subgroup (labeled B. in the table) and will be discussed later.

TABLE I.—ANALYSIS OF A TYPICAL CASE OF TETRACHLORETHANE POISONING

Date.	White blood cells in thousands.	Per cent.					Broken white blood cells per 100 formed white blood cells.	Red blood cells in millions.	Red cells.*				Platelets. †	Symptoms.‡
		Polynuclears.	Lymphocytes.	Large mononuclears.	Eosinophiles.	Basophiles.			Achromia.	Anisocytosis.	Poikilocytosis.	Polychromatic.		
Jan. 2	9.0	59	33	6	2	0	5	5.0	0	0	0	0	N	0
Feb. 18	8.5	56	31	13	0	0	4	..	..	..	..	..	..	0
Apr. 20	11.2	50	30	20	0	0	8	4.9	+	0	0	0	N	0
May 1	12.0	45	25	29	1	0	22	..	+	+	0	0	+	+°
May 13	15.6	39	20	40	1	0	31	4.7	+	+	+	+	+	++§
May 16	15.2	43	22	35	0	0	36	..	++	++	+	++	++	+++
May 23	11.4	45	29	25	1	0	18	4.7	..	..	..	..	..	0¶
June 6	10.6	50	38	12	0	0	7	4.7	+	+	0	0	+	0

* Red cells:
+ indicative of earliest recognizable changes: Very slight achromia, just distinguishable variation in size or shape; one polychromatophilic cell per 10 to 12 oil-immersion fields.
++ moderate achromia: Definite but slight variation in size and shape; one polychromatic cell per 4 or 5 fields.

† Platelets:
+ very slight increase in numbers.
++ moderate increase in numbers.
N = normal.

‡ Symptoms, degree indicated by + signs:
+ abnormal fatigue, discontent, constipation, headache.
++ a greater degree of above symptoms, coupled with insomnia and irritability.
+++ further increase of above symptoms, with addition of anorexia, nausea, vomiting and slight jaundice.

§ Taken off work.

¶ Jaundice still present; no subjective symptoms.

Group II consists of 23 male employees whose large mononuclear cells remained below 12 per cent. These men showed no symptoms

of poisoning in spite of exposure to tetrachlorethane to essentially the same degree as those placed in Group I.

TABLE II.—SUMMARY OF BLOOD FINDINGS IN SIXTY-EIGHT TETRACHLORETHANE WORKERS

	Number of cases.	White blood cells* in thousands.	Per cent.					Broken white blood cells per 100 formed white blood cells.	Red blood cells* in millions.	Red-cell abnormalities.†	Platelets.	Symptoms. ‡
			Polynuclears.	Lymphocytes.	Large mononuclears.	Eosinophiles.	Basophiles.					
Group I *a*	21	12.7	40.0	30.5	26.5	2.5	0.5	25	4.4	++	+	+
Group I *b*	4	10.2	41.0	35.5	22.0	1.0	0.5	8	4.9	0	+	0
Group II	23	10.3	62.5	27.0	8.0	1.5	1.0	5	5.0	0	N	0
Group III	20	10.2	60.0	29.0	9.0	1.5	0.5	5	4.3	+	N	0

The figures and data given represent an average obtained at the time the large mononuclear cells were highest or near their height in each instance.

Group I is composed of individuals whose large mononuclears were over 12 per cent.

(*a*) with symptoms due to tetrachlorethane.

(*b*) without symptoms due to tetrachlorethane.

Group II is composed of men and Group III of women whose large mononuclears were less than 12 per cent.

* Representative average counts; entire group not always included.

† + indicative of the very slightest definite red-cell abnormality, such as due to a very mild degree of secondary anemia from any cause, and as often found in the women employees.

++ indicates a rather greater degree of red-cell abnormality, such as referred to in the legend of Table I.

‡ + means the presence of symptoms due to tetrachlorethane.

0 means there were no such symptoms.

In Group III are put 20 women, whose large mononuclear cells were below 12 per cent and who had no symptoms of poisoning. They serve fairly well as a control group, as they had almost no exposure to the poison.

Group II and III require no further comment, as neither symptoms nor significant blood abnormalities occurred.

The pathological increase of the large mononuclear cells, together with other blood changes which occurred in the cases of Group I, are to be interpreted critically. The typical blood changes that occurred in the individuals placed in this group have been shown in

Table I. The typical case shows the large mononuclears rising progressively from approximately 10 to 40 per cent over a period of four to eight weeks, at which time preliminary symptoms of poisoning occur. Four cases developed over 40 per cent of large mononuclears, the highest count recorded being 56 per cent. The 9 more severely poisoned cases had usually, but not always, a slightly higher percentage of large mononuclears than the 12 very mildly poisoned workmen. In a typical case, though the symptoms of poisoning may progress over a period of a few days, there occurs no further increase of the large mononuclear cells. These cells gradually fell from the vicinity of 40 per cent to a level of about 12 per cent in three to six weeks, while if they rose to but about 25 per cent they fell in a shorter period of time. Their diminution is nearly always to be looked upon as a desirable sign, but perhaps this may not always be the case, as will be referred to later. Sixteen of the cases of Group I were of the typical sort. However, our data shows that there may be marked variation from the typical case which would appear to be somewhat dependent upon the given individual's resistance to the poison and to the amount of exposure to it. The worker may show the same progression of blood changes and symptoms, but develop the blood changes over a period of less than two weeks instead of over four to eight weeks. This is unusual and occurred in but one instance. Again, an individual may have over a period of months a relatively high level of his large mononuclears (10 to 20 per cent) and then suddenly symptoms will develop coincident with a rapid rise of the large mononuclears. This may occur, as it did in 1 case, without realizing that any increased exposure to the poison had occurred, or, as occurred in another case, a rapid rise from 10 to 40 per cent occurred, coincident with the acute development of symptoms due to increased exposure to the poison. In such an instance the symptoms may develop too rapidly to be foretold by the usual routine blood examination. However, such cases show the characteristic blood picture, which returns to normal in the usual manner.

From observations on 2 cases, developing an acute respiratory infection, it was apparent that an intercurrent infection might lower the individual's resistance to the poison. This was evidenced

by the fact that their mononuclears rose in about twenty-four hours from about 10 to approximately 40 per cent. Symptoms believed to be dependent upon the poison then developed which were sufficiently ill-defined to be plausibly explained as due to the infection, as similarly the blood picture might be interpreted. Coincident with removal from the poison, and with subsidence of the infection, the mononuclear count fell somewhat more rapidly than was usual in the typical case.

In spite of these variations from the typical case it is to be borne in mind that, as a rule, the insidious progressive onset of poisoning is the commonest form, the one to guard against and the one in which symptoms can be foretold from the blood changes.

The fact that the occurrence of clinical poisoning can often be foretold by the blood from something more than an increased percentage of large mononuclear cells must be borne in mind. It may be foretold by progressive increases of these cells, together with changes in their character and other blood features yet to be commented upon.

An increase of large mononuclears, on the average as great as occurred in the presence of symptoms, occurred in a few cases some months before symptoms of poisoning developed, though before and after the appearance of such symptoms there was a progressive increase of the cells as well as changes in their character. This indicates that any employee carrying an abnormally high mononuclear count is to be looked upon as potentially a poisoned case who may at any time develop clinical symptoms.

Four men, as previously noted, those of Group I, showed an increased percentage of large mononuclears without developing symptoms of poisoning. Two of these were men of about forty-five years of age, who had been exposed to the poison upward of a year and who showed an increased percentage of their large mononuclear cells for three months. The two others were boys of about seventeen years of age, who had been exposed to tetrachlorethane for about twenty-four to forty-eight hours. The increase of the large mononuclears in the older men may be explained on the ground of a constant chronic irritation by tetrachlorethane—a calling out of the first line of defense. This is further suggested

by the fact that when these men changed their work to a place of less exposure their large mononuclears became markedly reduced over a period of four months. The case of the boys, whose large mononuclear count fell nearly as rapidly as it rose, although they continued at work, we may easily assume as being due to a reaction to the substance. It would seem that as their bodies found they could functionate without the need of so sharp a reaction to tetrachlorethane the count fell.

A high mononuclear count appears to be the earliest sign of a reaction to tetrachlorethane. The mere fact that there is an increase of the large mononuclears is not sufficient evidence to advise that a man be employed in different work. A high count alone which does not become progressively higher has nowhere nearly the same significance as a count slowly rising, particularly over a relatively short period of time, except apparently when individuals are first exposed to the poison.

Besides the fact that a progressive increase of large mononuclears is more significant of a deleterious effect of tetrachlorethane than a stationary level of these cells there is another alteration in the large mononuclears that is significant in indicating the clinical prognosis. This is referable to their detailed character. It was found that there was a rather distinct relationship between the youth of the large mononuclears and the degree of clinical poisoning. With an increase of large mononuclears, the greater the number of young large mononuclear cells the sooner there developed clinical evidence of poisoning; while if symptoms were present and increased there occurred an increase in the number of young large mononuclears.

We consider that the younger large mononuclear cells are told from the older in the following ways: The nucleus of younger cells is oval or round and usually less homogeneous, in contrast to the indented or lobed, rather dense nucleus of the adult cell. The more youthful cells are usually of a different size than the more mature ones; they are sometimes smaller, with the cytoplasm relatively less in amount in proportion to the nucleus than in the adult cell; but, on the contrary, are usually larger with a relatively large amount of cytoplasm. It is characteristic for the cytoplasm of

the younger forms to be more basophilic and contain fewer azurophile granules. A certain amount of difficulty was experienced in making accurate differential counts in these cases, dependent upon the great variation in the character of these cells, as indicated above. At times differentiation from derivatives of the lymphoid system is perhaps impossible. The use of Goodpasture's modification of Graham's oxydase stain, which stains the granules of cells of myelogenous origin and not those of lymphocytes, was found of great assistance as a supplement to the Wright stain.

An increased fragility to mechanical injury (as from pressure of a cover-glass) of white cells may be looked upon as evidence of youth. Cells that have become broken cannot be classified in a differential count, but when one can trace a series of cells from those that are intact to those that are broken one may conclude that the majority of such broken cells are of the type to which they can be traced. In our preparations the majority of the broken or fragile white cells could be interpreted best as large mononuclears. The frequency of these disintegrated cells has been indicated in the tables. They were found quite consistently in increasingly greater numbers as the clinical symptoms appeared or progressed and were associated with increases of large mononuclear cells showing the other signs of youth. The increases of these fragile forms occurred with too great regularity to be considered purely due to careless technic, though of course a few such cells, some of which are probably adult cells, will occur in practically any blood smear.

The number of these distorted cells, together with the number of other youthful large mononuclears, is to be looked upon as a very definite factor in forming a prognosis. The more there were of such cells, indicating increased activity of the tissue from which they are formed, the sooner definite symptoms occurred or more serious symptoms developed. It was common to find three to seven days before symptoms developed 10 to 20 broken cells to 100 formed white cells. With the presence of definite symptoms they increased on the average to about 40 per 100 formed white cells, while in some of the more marked and more acute cases of poisoning they reached to over 100 per 100 formed white cells.

No case of poisoning showed less than 15 fragile cells to 100 formed white cells. Likewise, the formed large mononuclears in the poisoned individual had very varying characters: some were very young and others mature, with all variations between these extremes. In contrast to this picture those 4 cases which did not develop symptoms and those poisoned cases which had high large mononuclear counts, some weeks or even months before poisoning occurred, showed at such times usually but about 10 fragile to 100 formed white cells. There was also but little variation in the character of their mononuclears, which were particularly of the more adult forms. In other words, when symptoms of poisoning occurred there was much greater activity of formation of the large mononuclears than when symptoms did not occur, even though they might be increased in numbers, indicating a reaction to tetrachlorethane.

It is evident that increases of the large mononuclear cells indicate a reaction to tetrachlorethane and appear to be an early sign of poisoning which may occur before clinical symptoms are apparent; while the youth of these cells, as told by their histological appearance, is a sign that indicates somewhat the degree of poisoning. These signs are of considerable importance in deciding whether a man should be laid off from work exposing him to tetrachlorethane, and, if so, for how long. Before formulating rules as to how blood examinations may be used to prevent poisoning and for its diagnosis, it is desirable to describe the other less important alterations of the blood that were observed in these workers. These other blood changes are referable to the total white count, further alteration in the differential white count, the red cells and platelets.

Accompanying the increase of large mononuclears there occurred a slight absolute increase of the white cells, never above 22,000 per c.mm. and usually about 12,000 per c.mm. The higher counts were liable to be found coincident with the greater increases of the large mononuclears and when they showed the greater histological changes from normal. Among 23 individuals exposed to tetrachlorethane, but presenting no signs or symptoms of poisoning, and who had no other blood changes, the white count averaged 10,300. These observations suggest that anyone who comes in

contact with tetrachlorethane generally has a slightly elevated white count, and thus, that this chemical tends to cause an increase of the total number of white cells.

Alteration in the differential count other than alteration in the large mononuclears is of relatively slight importance. Often there was found an initial relative decrease of the polynuclears, without particular fluctuation of the lymphocytes, coincident with the increase of large mononuclears. With a decrease in the percentage of large mononuclears the lymphocytes frequently rose more rapidly than did the polynuclears, often reaching a percentage higher than that preceding the initial changes due to tetrachlorethane. It is our impression that should a case become distinctly severely poisoned (one example of which we have seen, but who was not in the group of workers we have especially studied) the large mononuclear cells may fall, the defensive reaction having been overcome, and one may then find a definite lymphocytosis. Thus if the clinical symptoms of poisoning were progressive, and the large mononuclear count was falling while the lymphocytes were rising, it should not be necessarily interpreted that the individual was getting better, but that he was getting worse. In morphology the lymphocytes seemed to be for the most part normal. In some cases, particularly with an increase in their numbers, there was a somewhat increased percentage of large lymphocytes, some of which were atypical.

Besides alterations in the percentage of polynuclear neutrophils referred to above the only other abnormality they presented was that at times young forms were found in the peripheral blood. Young forms in rather small numbers were especially seen when there was evidence of considerable heightened activity of the formation of large mononuclears.

No definite variation normal was found in the polynuclear eosinophils or basophils.

The abnormalities of the red cells that were observed were distinctly of secondary interest and importance as compared with the white cells. The average red count in 11 men (25 counts) with symptoms of poisoning was 4,400,000 (varying from 3,900,000 to 5,200,000), while in 10 men (10 counts) who had no increase of

their large mononuclears, and who had no symptoms, the average red count was 4,900,000 (varying from 4,700,00 to 5,200,000). The average count among 9 women (9 counts) who had no symptoms of poisoning, and who for the most part were typically pale factory girls, was 4,300,00 (varying from 4,100,000 to 4,600,000). No accurate hemoglobin determinations were made. By the Tallquist scale it was apparent that there was a slight reduction of hemoglobin that roughly paralleled the red counts. Abnormalities of the histology of the red cells in the poisoned cases were more evident than the variation from normal of the red count and hemoglobin. The amount of achromia, variation in size and shape and polychromatophilia, that was observed in the average case has been indicated in the tables. There was no evidence of abnormal fragmentation of the red cells, such as occurs in trinitrotoluene poisoning, to suggest that any important degree of blood destruction occurred in tetrachlorethane poisoning. In no case were the abnormalities of the red cells present to more than a slight yet definite degree. The abnormalities were distinctly more apparent in the men developing poisoning whose red counts in turn were lower than in the men who did not develop poisoning, yet higher than in the women who were not poisoned.

All the instances of this simple anemia unexplained by some other cause than due to tetrachlorethane, and as particularly told by the histology of the red cells, developed in cases that showed increases of large mononuclears or clinical symptoms of poisoning. However, every case of poisoning did not develop anemia. The red-cell changes were somewhat more marked in the more chronic and more severely poisoned cases. From our data it appears that the anemia is slowly progressive, especially when symptoms persist or with recurrence of poisoning, but it does not become serious. In any given case the amount of red-cell abnormality alone would be of very slight importance in judging the state of the worker. However, the presence of red-cell abnormality is to be looked upon as an undesirable finding, and, particularly when present with the more important white-cell abnormalities, is to be looked upon as an additional sign to indicate that the worker should be removed from the poison. It is also probably wise to

consider that the poisoned individual with anemia should remain longer from exposure to tetrachlorethane than one without anemia, and that individuals with anemia should not be exposed to this substance.

The blood platelets, as estimated from fixed smears, were frequently found slightly increased above normal. As might be expected, their increase was usually more apparent in those cases showing distinct abnormalities of the large mononuclears and changes in the red-cells than in those with the lesser changes in these elements. Abnormally large platelets were rather frequently found in cases showing other abnormal blood histology.

From this study of the blood of tetrachlorethane workers the hopes that blood examination might be of value in regulating the employment of those exposed to this poison have been rather definitely fulfilled.

On the basis of the correlated study of the clinical picture and the blood the following recommendations regarding blood examination are made for the prevention and diagnosis of poisoning by tetrachlorethane. These recommendations apply when the substance is used under such satisfactory conditions as were observed during our investigation:

A routine blood examination should be made before an individual is accepted for work. This will particularly establish his normal level of large mononuclear cells and will determine the presence or absence of any anemia. As anemia probably favors the development of poisoning, if there is any definite degree of it the individual had best be rejected:

A critical examination of a stained blood smear should be made on each new employee at the end of the first and second week and then once every three or four weeks during the first six months of his exposure to tetrachlorethane. After the first six months less frequent examination of the blood is necessary. It appears that a given individual is much more likely to develop evidence of poisoning during the first six months of his exposure. This is substantiated by the fact that in the individuals examined 17 cases of poisoning occurred among the men employed for less than six months, while but 4 cases occurred in a group of essentially the

same number who had been employed for over six months. A new employee may have an initial reaction to the poison. In the event of his being distinctly susceptible the blood picture may progress from that point. In the event of his being less susceptible his initial mononuclear rise subsides and probably some tolerance is established.

If the routine blood examination reveals the large mononuclear cells above 12 per cent, more frequent examination is indicated. The frequency of such examinations will depend upon the relative rate of increase of the large mononuclear cells, together with their numbers showing signs of youth, as well as the presence of even the most trifling clinical symptoms.

Any individual with a percentage of large mononuclear cells of over 12 must be considered, for the purposes of diagnosis and prognosis, potentially as a poison case, because an increase of the large mononuclear cells seems to be the first sign of a reaction to tetrachlorethane. However, this arbitrary figure of 12 per cent cannot be utilized alone in regulating his employment except ideally and impracticably. From the point of view of the employer such regulation would result in serious economic loss and from the outlook of the employee injustice would be done. Also it is very evident that no harm comes to the employee who is allowed to reach the point of developing the very earliest clinical symptoms of poisoning.

We have already noted that the large mononuclears in the 12 mildest cases were often as high as in the 9 more severe cases who were removed from work. However, in these more severe cases the presence of larger numbers of younger mononuclear cells and broken cells was very much more constant than in the 12 milder cases. The determining factor in the removal of the 9 men was not only that the blood changes were more marked but that the clinical symptoms were more marked. In both instances it was on account of the blood examination that the cases came under critical observation; some, however, were allowed to develop a greater degree of poisoning than we now feel is desirable to permit. Of the 9 severer cases all but the 2 acute ones could have been removed from work sooner than they were, and should have been, from the evidence of poisoning obtained from the blood as well as by symptoms.

The regulation of employment, therefore, from the point of view of the blood findings has to be based particularly both on the relative increase of the large mononuclear cells over a definite period of time and upon the number of the young and fragile forms.

It may be stated that a progressive increase of large mononuclear cells alone does not call for removal of an individual from work but calls for his close observation. If the slightest clinical symptoms develop it is desirable to remove him from exposure. It is permissible, however, to allow him to continue at work for a few days unless his symptoms increase in the least or his blood picture becomes more abnormal, as particularly evidenced by an increase of the formed and broken young large mononuclears.

Any case having symptoms, even if very slight, for more than a few days, with abnormalities of the blood, should be excused from work. An increase of large mononuclear cells from 12 to 20 per cent, or from 20 to 40 per cent, over a period of two or three weeks, is to be looked upon as of much more significance than a similar increase over a period of two or three months.

In the same way that the blood serves as an important guide to indicate when to take an employee off work, so it serves in regulating his return. No individual should return as long as any clinical symptoms persist, with the possible exception of a very slight and subsiding jaundice. If the subjective symptoms have subsided and the large mononuclear cells have been reduced by at least 50 per cent from their highest point, then, even perhaps in the presence of slight jaundice, if none of the younger forms occur in the blood, a man may be allowed to return to work provided he be kept under the strictest observation. The absolute percentage value of the large mononuclear cells is of less importance; that is, it does not appear to be absolutely necessary for the large mononuclear cells to return to 12 per cent or less before allowing an individual to resume work. The presence of definite anemia is a contraindication to returning to work, and is usually sufficient evidence to advise a change of occupation.

The period of time that the poisoned cases were removed from exposure varied from a matter of a few days to six or even eight weeks. In general the sooner a case was removed from work after

definite blood changes and slight symptoms occurred the shorter was the time lost.

The reason that an individual should be watched particularly carefully upon returning to work after poisoning is because there seems to be a tendency for those who have become poisoned once to become so again, and usually relatively more rapidly than before. If an individual becomes poisoned twice within a few weeks, or several times some months apart, it is wise that he should change his employment. It was found that with the recurrence of poisoning the blood changes were usually more severe and more rapid.* As alluded to above it may be desirable to supplement the routine blood examination with others, according to the exigencies of the case. These more frequent tests may be made purely because the blood has been abnormal or because some symptoms have occurred. It is to be clearly understood that every employee developing any symptoms, no matter how trivial, which could in any way be interpreted as due to tetrachlorethane should have a critical blood examination. Under such conditions the blood will materially aid in the differential diagnosis of the cause of the symptoms. If the typical blood changes described as due to tetrachlorethane are present the symptoms had best be credited to the poison, while if no increase of large mononuclears occurs the symptoms can be attributed to some other cause. In this connection it is desirable to note that occasionally certain acute infections and other conditions may cause a considerable increase of the large mononuclears. However, confusion in the diagnosis between tetrachlorethane poisoning and conditions that occasionally give a blood picture simulating that due to tetrachlorethane must be rare.

We feel that blood examination utilized in the manner outlined above is the most important single criterion that we have for indicating the onset of poisoning by tetrachlorethane or its initial symptoms. It is concrete and the important changes that take place are constant. This is in contrast to the very earliest symptoms of poisoning which are vague and indefinite. Such symptoms,

* Instances of recurrent poisoning are not included in the previously referred to 21 cases of poisoning.

however, are shown to be quite definitely due to poisoning when present with the blood changes described. During our study we have been able to prophesy frequently from the blood some days before clinical symptoms occurred that such symptoms would appear. The blood changes were found to be invariably the first symptoms or signs to appear, and other than jaundice were the last to disappear. However, blood examination is not infallible in detecting poisoning before clinical symptoms develop. This is because occasionally, from undue exposure, symptoms may develop so rapidly that they could not be anticipated from routine blood examinations.

The acute development of even slight symptoms may be due to a relatively sudden greater exposure to the poison or to the action of some factor not clearly understood, as a lowered resistance from infection. However, such cases will show the typical blood changes, so that diagnostically the blood is always helpful.

One should most emphatically not rely on blood examinations alone to keep the health of these employees at its best. It aids to detect the early, often apparently insignificant, symptoms due to poisoning and serves as an absolute check upon them. Blood examination should be utilized as a valuable method to aid in the prevention of serious poisoning and in the diagnosis and prognosis of poisoning. However, proper clinical supervision of those exposed to tetrachlorethane, as outlined by Dr. Parmenter, is of the utmost importance. If blood examination is used intelligently it can aid to reduce materially the time lost from work, for early removal from exposure may reduce the time out to less than a week, while cases that go on to the development of jaundice usually require at least a month.

It is only by careful observation, at least every few days, together with a clear conception of the initial symptomatology of the poisoning, and with utilization and proper interpretation of blood examinations, that poisoning can be kept at a minimum. Under such circumstances, carried out by Drs. Parmenter and Smith, the number of working days lost, due to poisoning, in this artificial silk plant has been reduced for the five months preceding March 1, 1921, by about 40 per cent below what the number was for the five

months beginning January 1, 1920, when the original investigation reported here was conducted.

SUMMARY. 1. A study of the blood of 63 individuals exposed to a greater or lesser degree to tetrachlorethane indicates that blood examination is of value in the prevention of tetrachlorethane poisoning and in the diagnosis and prognosis of poisoning by this substance.

2. All cases presenting clinical symptoms of tetrachlorethane poisoning (21 in number) showed characteristic blood abnormalities. The blood changes usually can be observed before clinical symptoms develop.

3. The blood abnormalities include:

(*a*) A progressive increase of large mononuclear cells, often reaching 40 per cent. This is the most important change.

(*b*) The appearance of many immature large mononuclears.

(*c*) A slight elevation in the white count.

(*d*) A progressive but slight anemia.

(*e*) A slight increase in the number of platelets.

4. A percentage of large mononuclear white cells, about 12, is the first sign of a reaction to tetrachlorethane and is a signal for close observation of that individual. All individuals with such a picture do not necessarily develop clinical symptoms of poisoning.

5. The presence of a considerable number of young large mononuclear cells, some formed and many broken, is to be looked upon as indicating a severer condition than when a case has the same number of more mature large mononuclears.

6. A consideration has been given to the utilization of blood examinations in connection with careful clinical observations in the regulation of employment of those exposed to tetrachlorethane.

AN ACCURATE METHOD FOR THE CLINICAL DETERMINATION OF EARLY ARTERIAL DISEASE

By HARLOW BROOKS, M.D.
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The diagnosis of arterial disease must be made early if it is to be utilized to any definite practical end in clinical cases. By the older methods early diagnosis is rarely or never possible except largely on supposition or on historic grounds. As a result very little curative effect is accomplished in cases of arteriosclerosis except in early specific cases in which, as a rule, the diagnosis of arterial disease is based upon the law of probability rather than on actual detection of changes in the bloodvessels.

Thus far early arterial disease has been detected chiefly by an ophthalmoscopic study of the retinal arterioles. This has indeed proved most valuable, but its successful employment presupposes a considerable technical skill with the ophthalmoscope, and a very wide experience is necessary for accuracy. The method at its best is hardly applicable for routine use in the hands of the average clinician, and yet the data which its utilization furnishes is material of the most elemental importance in the subsequent treatment of any case.

If real results are to be expected in the management of this type of disease it must be instituted very early, and, as a rule, this is not possible or probable under usual diagnostic routine or until such time as irradicable lesions have been inflicted.

It is generally supposed that the retinal arteries are among the very first to show indications of developing arterial disease, and this is in all probability a correct statement; but we must recall that in very many cases, especially in arteriosclerosis, lesions are not always

diffuse or universal, but are more or less limited to or more pronounced in certain systems or distributions. Even if this assumption be accepted it still remains that the retinal vessels correctly typify those of the most primitively important centers, that is, those of the cerebrospinal axis.

The method to which I wish to call your attention was originally demonstrated to me by Dr. David Dennis, of Erie, Pennsylvania, who already has made several contributions to medical literature concerning the method, chiefly in ophthalmologic journals.

It is based on the fact that the arterioles and other conjunctival bloodvessels show changes quite consonant with those present in the retinæ. That this is a justifiable assumption is borne out by the fact that the conjunctival arterioles originate from the anterior and long ciliary branches of the ophthalmic artery and to a lesser extent from the external and internal palpebral branches. They thus represent in a very typical way the circulatory paths within the brain, quite as much so at least as the retinal arteries which spring from the same trunk.

Study of the vessels is accomplished by the use of two very simple and easily manipulated instruments which are usually in the pocket of the average practitioner. The ordinary pocket electric flashlight, of which the most convenient for this purpose is the "fountain-pen" type, is used for illumination. The patient is directed to turn his eyes to either the one side or the other, and the light, held at a distance of about 3 or 4 cm., is directed obliquely on to the ocular conjunctiva. Study of the vessels is then made through an ordinary ophthalmologist's loop, which is the most adaptable to the purpose, although other lenses are also fairly satisfactory. The loop is held at the proper focal distance, and for most satisfactory study the eye of the observer is brought close to the lens, just as in the use of the microscope. The vessels under study in the various levels of the membrane are brought sharply into focus by moving the lens to and fro, and for the purpose of steadying it the fingers of the lens hand may be rested on the orbital arch of the patient. The study may be made in the diffuse light of the examining rooms or even more satisfactorily in the dark room. When my flashlight has been too weak I have secured my lighting by the use of the ordinary head

mirror and the usual examining lamp, such as one uses in the examination of the nose and throat. A sharply condensing light, such as is employed in the illumination of the sinuses and so on, is of course very desirable, but the ordinary small electric flashlight is all that is needed.

One is thus able to study in a most satisfactory way the larger branches of the ciliaries and palpebrals, the tiny arborizing arterioles and even the capillary circulation in the occular conjunctivæ as well as the return circulation in the venules and the larger ciliary and palpebral branches of the ophthalmic. These vessels are displayed, as it were, beautifully spread out on an ideal pearly white background which forms a most pleasing contrast to the yellow red of the arterioles and the deeper carmine color of the venules. The arrangement for study is as definite and satisfactory as is possible in a well-stained and technically perfect microscopic slide, and by slight variations in focus a vessel may be followed throughout its entire arborization. The method is much more satisfactory than the study of the retinal circulation because of the sharp contrast of the white background as compared to the pinkish tinge of the retina, and it is much more representative of the internal cerebral circulatory condition than the tiny retinal artery, since we have here represented for our study four systems of arteries all derived from the same branch of the internal carotid, namely, from the ophthalmic. A further great advantage over the study of the retinal artery is represented in that we are here able to observe in the conjunctival circulation the effects of arterial disease on the adjacent tissue. This is particularly fascinating in cases of arteriocapillary fibrosis, in calcifications and fatty degenerations and in instances of hypertension of marked degree. A disadvantage of the method lies in the fact that in cases in which disease of the conjunctiva is present, local vascular lesions are found, particularly in conjunctivitis and in the membrane of patients who have been exposed to traumatism of wind, intense light and of irritating chemicals and gases.

The studies of Dr. Dennis, of Erie, and of Dr. Reese, of New York, have shown quite finally that these changes in the conjunctival vessels are entirely comparable in any given instance with

the alterations manifest in the retinal arteries; this I have also corroborated within the past four years that I have been using the method as a routine, in so far as my skill with the ophthalmoscope may be trusted.

The great advantage to the clinician in the method is that a highly sufficient technical skill may be acquired with a few days' practice. It demands no special instruments and less time is required for the intimate study of the minute circulatory changes in the cerebral vessels than is necessary for a reasonably careful palpation of the radial, brachial or temporal arteries. There is no longer any excuse for mere speculation as to the condition of the bloodvessels of the brain; from 30 to 120 seconds of time is sufficient to give one a very satisfactory and entirely accurate knowledge of such changes if present.

I have found the study most helpful and of tremendous advantage in the detection of the earlier changes present, as in syphilis and arteritis, from any cause. I have become particularly interested in the study of the terminal arteriole changes in hypertension.

One of the pitfalls into which you will sooner or later fall if you follow this method is the employment of the corneal microscope, by means of which the most intimate study may be made not only of the microscopic changes in the walls of the arterioles, capillaries and venules, but also the physiologic and pathologic variations in the circulating stream can be made out with greater lucidity than is possible in the web of the frog's foot under the low powers of the microscope. This requires, as I can abundantly testify, a very considerable degree of technical skill, costly and delicate instruments, and most expensive of all a very large amount of time and patience. This means of study of the conjunctival circulation, however, is indispensable to check up, as it were, the gross pathology observable with the hand lens. The short distance which I have already traveled in the art shows me that it offers a most valuable method for the microscopic study of the circulatory conditions of the brain. This is transferable by inference to the other parts of the body in general arterial disease. It is a study which bids fair to be of even greater value to the internist than to the ophthalmologist.

The most signal benefit conferred by the methods presented is,

of course, the ready detection of the very early changes in the arterioles, which may lead on to generalized arterial lesion, which plays so important a role in the evolution of many internal diseases. By these studies one may detect sufficiently early changes in the arterial system to permit him to inaugurate modifications in habits of life, exercise, diet, etc., also to render possible, in association with proper medication, the introduction of a treatment sufficiently early so that real preventive measures are attainable in many cases.

I have used the study of the conjunctival vessels as a routine in my physical examination of office cases and to a considerable degree also on my hospital services now for about three years. I have been able to satisfy myself that the conjunctival vessels very satisfactorily represent the existence and type of a general arteriosclerosis, as confirmed finally by the usual methods of study.

Changes detectable in the conjunctival vessels admit of practically the same classification which we recognize pathologically in our study of anatomic material, and a little technical experience readily permits the early recognition of these changes, as it were, *in vivo*.

I wish to present here very briefly a study which I have made of the conjunctival vessels in cases of hypertension. One of the most striking changes which the observer notes in cases of hypertension is the elevation of the more superficial vessels above the surface of the conjunctiva, so that when the light is divided diagonally on the surface of the conjunctiva a reticular framework made up of the tense bloodvessels is shown. The arterioles show in early phases also a clear sharp line of demarcation, sharply differentiated off from the surrounding tissues as though overtensely filled. This is manifest even when there are extensive degenerative infiltratory or inflammatory alterations in the surrounding tissues.

Many of the more superficial vessels are thrown into numerous convolutions, as is seen grossly in the contorted arteries and the calcifications of old age. It must be noted, however, that the venules of the conjunctivæ are often markedly convoluted and that this is apparently a normal condition in certain persons, for I have observed its frequent presence in the conjunctival veins of infants and youths entirely independent of any apparent circulatory disease.

This is not, however, true of the arteries in which convolutions apparently bespeak only a disease process associated with arteriosclerotic changes or with hypertension.

When a period of hypertension of long standing has been succeeded by a hypotension, as from a circulatory decompensation, this elevation and contortion of the arterioles is still preserved and may serve as a sign apparently that a hypertensive condition has preexisted.

Another very frequent but by no means constant finding in the conjunctival vessels in hypertensive cases is the existence of nodes of lumen contraction, so that though the vessel walls are represented by a relatively broad yellowish or whitish streak but a mere trickle of golden red represents the contracted lumen, at the proximal end of which a minute dilatation of caliber is demonstrable. This is particularly evident in very long-standing cases, and it apparently represents an arteritis or arteriofibrosis with obliteration of the lumen. The extreme stage of this alteration is represented by complete obliteration, so that vessels are represented merely by yellowish or grayish cord-like structures. Where many of these obliterated vessels are present in the conjunctiva the whole membrane is given a grayish or yellowish-white color, so that an appearance of extreme anemia may be presented while the blood picture shows a normal condition of affairs quite surprising to the clinician who is accustomed to judge as to the degree of anemia from the color imparted by the conjunctival vessels. This statement comprehends the palpebral as well as the ocular conjunctiva.

The first two signs of hypertension appear relatively early in cases of hypertension, the latter only in the long-standing cases.

Tiny aneurysmal dilatations of the conjunctival arterioles are not commonly seen in hypertensive cases. They are, as Dennis has stated to be the case, most common in instances of true arteritis where local changes in or about the arterioles predominate, but of course they may, on theoretical grounds, be expected in those cases in which an arteriocapillary fibrosis is the cause of the hypertension.

DISCUSSION

Dr. S. B. Wolbach, Boston, Mass.: What was the caliber of these vessels?

Dr. Harlow Brooks (closing): Some are very narrow and some very broad. The size varies as much as does that of the radial artery, but one will not have any difficulty in distinguishing pathologic vessels.

FURTHER STUDY OF THE QUANTITATIVE VARIATIONS IN THE VIBRATION SENSATION

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The first contribution to the study of the vibration sensation was made by Rumpf in 1889. A steel tuning-fork was used and the handle was so made that it was suitable for application of it to the skin surface after the vibration was started. Forks of different rate of vibration were used in this study, and it was found that in different parts of the body different rates of vibration were better appreciated. Rumpf regarded the procedure merely as a means for the more careful study of skin sensation.

In 1907 Treitel, unaware of Rumpf's work, experimented with a tuning-fork of 128 vibrations, with which the variation in the length of time of the perception of the sensation was studied. He discovered the important fact that under certain pathologic conditions tactile sensation and vibration sensation bear a different relation to each other, and the one may be altered without alteration of the other. Contrary to the conclusion of Rumpf, he decided that the vibration sensation is to be distinguished from a sense of pressure and a sense of touch.

Max Egger[1] is usually regarded as the first to study the vibration sensation. Certainly, he was the first to demonstrate the feasibility of testing the sensibility of bones by means of a tuning-fork. The idea was suggested by the extreme sensitiveness of periosteal lesions. By lifting up a fold of skin in a thin subject and placing it between

[1] Jour. de physiol. et de path. gén., 1899, No. 3, vol. i; Rev. neurol., Paris, 1908, xvi, 345–351; Compt. rend. de la Soc. de biol., May 27, 1899, p. 423; Compt. rend. de la Soc. de neurol., February 4, 1904. Goldscheider: Berl. klin. Wchnschr., 1904, No. 14.

a vibrating tuning-fork and a fixed object only the very slightest vibration was detected. When the skin was allowed to fall back into place and the instrument was applied it was found that the appreciation was increased tenfold.

Egger demonstrated that this perception of vibration sensation was entirely independent of the presence or absence of cutaneous sensibility. More recently Goldscheider showed that Egger was in error in his conclusion that the vibration sensation is perceived only through bone. He found that the soft parts are susceptible, although not to so marked a degree owing to the physical property of the tissue. Goldscheider further thought that the observation was dependent in part on the condition of cutaneous sensibility. He experimented with anesthetization of the skin. To test the cutaneous sensibility the instrument was very lightly applied to the skin surface, and to test vibration it was applied much more firmly through the compressed skin. He did not regard it as a specific sensation but as an expression of the sensation of rhythmic interrupted irritation of the nerves that are responsible for the sensation of contact or pressure.

Bing is quite emphatic in stating that through cocain anesthesia tactile, pain and temperature sense may be abolished while the vibration sensation is preserved, and further that it is usually not noticeably decreased. He shows many good grounds for the sharp separation of tactile and vibration sensation.

L. F. Barker[1] is disposed to regard the sensation of vibration as due to the result of the stimulation of the touchpoints in the skin. He says that though this sensation may disappear before there is complete tactile anesthesia, still it seems to indicate that among the earlier disturbances of tactile sensation must be counted an inability to perceive successive impulses of short duration as separate sensations.

In 1903 Rydel and Seiffer[2] gave to the vibration sensation the name pallasthesia, which was a step in the direction of separating it as a different sensation from tactile and other sensations.

[1] Monographic Medicine, xiv, 136.

[2] Arch. f. Psychiat., Berlin, 1903, xxxvii, 488, 536; Neurol. Centralbl., 1903, No. 7; Berl. Ges. f. Psych. u. Nervenkr., March 9, 1903.

For clinical purposes Bing preferred to use the Gradenigo vibrator. A clear description of this instrument is not available. It seems to depend on an optical device as the means of determining a point of fixed amplitude. Barker preferred the C fork of 128 vibrations provided with a Gradenigo triangle on one prong, which permits, he says, of the application of vibrations of very different numbers, and the number at the instant of vibration disappearance may be exactly recorded. During the progress of this work various instruments with the Gradenigo triangle were used, but the results did not seem to justify a continuance because of the difficulty in general standardization. It seems justifiable to recall to the reader that a tuning-fork can only vibrate at a fixed rate regardless of the violence of the starting blow. The only thing the blow can alter according to its degree is the amplitude of the vibration or the "degree of swing." To reach some means whereby the amplitude can be standardized is ever the difficulty in perfecting an instrument for quantitative work.

R. T. Williamson,[1] the English pioneer in the study of the vibration sensation in neurology, mentions in his writings the advantages of the Gradenigo instrument. He was impressed with the fact that by it sensation in different parts of the body can be accurately measured. Williamson worked with a C^1 fork 9 inches long with prongs $5\frac{1}{2}$ inches long.

In 1911 J. L. M. Symns,[2] working in Guy's Hospital, adopted a tuning-fork for quantitative work which made possible the exact charting in numerical ratio of the responses of the subject at different points on the body surface. The instrument of Symns has a small "window" placed between the prongs at the free ends. If the amplitude is wide enough the prongs separate sufficiently to allow to be seen the two little slots, which differ slightly in depth. As soon as the vibration dies out sufficiently the window or slot is covered up. Unless the vibration is quite active the window will not appear. The observer strikes the instrument and then holds it up between his eye and a bright light, preferably a window, and

[1] British Med. Jour., July 20, 1907.

[2] British Med. Jour., 1912, i, 539; Guy's Hospital Reports, 1912, lxvi, 120; Quart. Jour. Med., 1917, ii, 33–58. Wood, E. J.: Guy's Hospital Reports, January, 1921.

continues to watch the gradually lessening light which is allowed to pass through the slot. At the instant when no more light can be detected the handle of the instrument is applied to the bony point under consideration and the patient told to note the vibration carefully, and at the instant when the vibration is no longer detected to give a suitable signal. The time during which the sensation is felt is recorded in seconds. Symns used a stop-watch, but it is not essential and has the disadvantage of requiring an assistant.

The present work was planned to serve as a continuance of the observations begun by Symns in the same clinic in 1911. Dr. Hurst as well as Dr. Symns were of the opinion that the normal range established by the latter had been based on too small a number of cases. One hundred normal individuals were studied in making a graph of the normal variations. The subjects were bed patients suffering from no nervous disease, willing to coöperate in the test and mentally intelligent. The graph constructed on this series of normal observations varied markedly from that constructed by Symns on a fewer number of observations. This was found to be due to the difference in the two instruments used. Through the help of Mr. H. G. Drew, of Messrs. Down Brothers, Ltd., of London, the makers of the instrument, it was soon learned that while the instrument used in the present work and that used by Symns were both of 108.5 double vibrations per second, and with identical windows, the sources of difference could be readily accounted for and in large measure eliminated in future manufacture. Variations in weight and temper of the shaft caused a variation in vibration appreciation. A vibration of 2 mm. swing at the fork end is differently transmitted at the handle end if the rigidity of the fork varies. The instrument used in this work has been carefully weighed and measured by the manufacturer and made the standard. The window, which is really a double slot, has been carefully registered and every precaution will be taken to manufacture instruments conforming to it. It must be remembered, however, that a hand-wrought instrument can never be exactly duplicated, and it is not possible to exactly reproduce temper. It was thought desirable to standardize the instruments in such a way that a normal graph might be supplied with each instrument to facilitate the exact recording of the curve in each individual case (Fig. 1).

In the diagrams it will be noted that the observations are made on various bony points of the body and recorded in seconds. The investigation is done in many other ways by a large number of observers. Dr. Henry Head does not use a windowed instrument and does not select the bony points used in the present work. One important part of his examination is timing the period during which the vibration can be detected on one side after the vibration can no longer be detected on the other side. That is, he holds the vibrator in contact with a part on the right side until the vibration cannot be felt, then quickly transfers the instrument to the other symmetri-

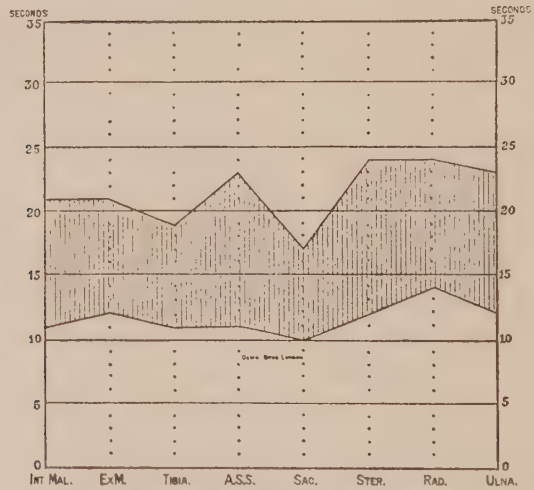

FIG. 1.—Normal range established in 100 non-neurological cases.

cal part on the left side and notes the time in which the lessening vibration can be felt. The procedure is then reversed. Dr. Gordon Holmes uses a simple heavy tuning-fork without windows and does not make time observations with standardized amplitude. His conclusions are based more on qualitative returns than on quantitative.

It was thought by those interested that the standardization of an instrument with a standard normal range, as has been attempted in this work, might open the way for numerous observations by many observers, resulting in the accumulation of data which is much needed

before final acceptance of many of the claims to be made can be expected.

The justification for this present work is the fact that until quantitative variations are appreciated the value of the test is lost at a time when its aid is most needed. Before total loss of vibration appreciation occurs there is a gradual dimming which with this instrument can be exactly recorded day by day and the progress of the process noted. For example, in tabes dorsalis there occurs late in the disease frequently an entire loss of vibration appreciation in the lower half of the body, but, on the other hand, early in tabes, before other signs of recognized value appear, it will be found that there is a definite quantitative diminution of the time during which these vibrations of standard amplitude are felt.

It is beyond the scope of this work to enter into a discussion of the nature of the vibration sensation and its relation to other sensations. Suffice it to say that under certain pathologic conditions of the nervous system there is definite departure from the normal.

The most notable variation of a fixed sort was seen in tabes, as has been mentioned. Eighty cases were studied in various stages, and in large part these cases were kept under observation for long enough periods to watch changes of vibration which occur much more markedly than of any other sensation. In not one of this series was there an exception to the rule that in the lower half of the body there always occurs a reduction in the time of appreciation of vibrations of a fixed amplitude. This reduction in time was singularly constant in its occurrence over the sacrum. It is rare, indeed, in any case of tabes, however early to record a time of more than five seconds over this point. In a large proportion of the cases there was no response at all over this portion of the body. In two or three instances the time recorded was as much as six or even seven seconds. Fig. 2 shows the curves of four cases of tabes overlaid. The general tendency to dip at the sacrum is well illustrated. In many instances this low response or absence of response occurs when the instrument is applied to the anterior-superior spine of the ilium. It is exceptional for the curve in tabes to fail to fall within the normal range in the upper extremity, and in some

instances there is really a high response in this part of the body. This is one of the distinctive differences between tabes and multiple neuritis. In the latter the normal range is seldom encroached on at any point and the "sacral dip" is absent.

This study has led to the conclusion that loss of vibration appreciation is the first sensory loss in tabes. This is illustrated in a man studied in Addison ward, Guy's Hospital, whose diagnosis was thoracic aneurysm. The blood in this case was Wassermann negative while the spinal fluid was strongly positive. There were no disturbances of tendon reflexes, the pupils were negative and

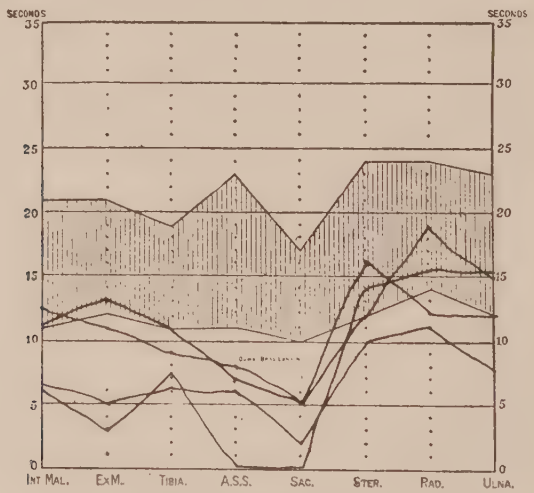

FIG. 2.—Vibration response in 4 cases of tabes.

careful study of skin sensation failed to reveal any areas of analgesia. The curve represented in Fig. 3 shows a low vibration response. It will be noted that the loss is not total but merely a quantitative change, which would be readily overlooked were the investigation merely qualitative. This patient was held in the hospital over a long period and we had the satisfaction before his discharge of seeing the development of areas of analgesia over the tibia. It seemed justifiable, after a study of a number of such cases, to conclude that analgesia followed the alteration of vibration response. In many instances the finding of a low vibration response prompted a careful

study of the whole body surface, with the result that certain areas of analgesia were sometimes found. In Fig. 4 the patient, whose reflexes were preserved, showed no appreciation of pain when a pin

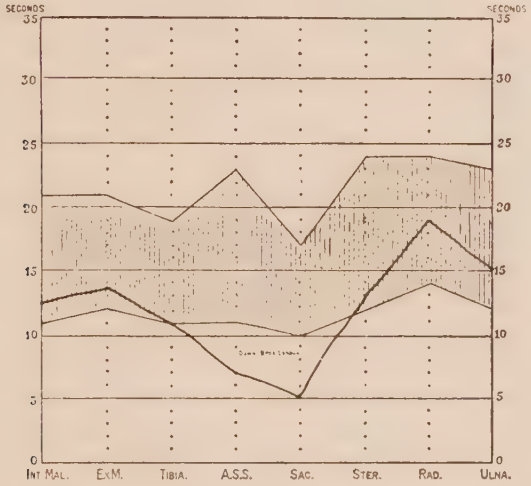

FIG. 3.—Man, aged fifty-five years. Admitted to Guy's Hospital (Addison Ward) for aneurysm. The blood was Wassermann negative but the spinal fluid strongly positive. The vibration curve was that of tabes and he was kept in hospital under observation until other signs developed sufficient for a confirmation of this diagnosis.

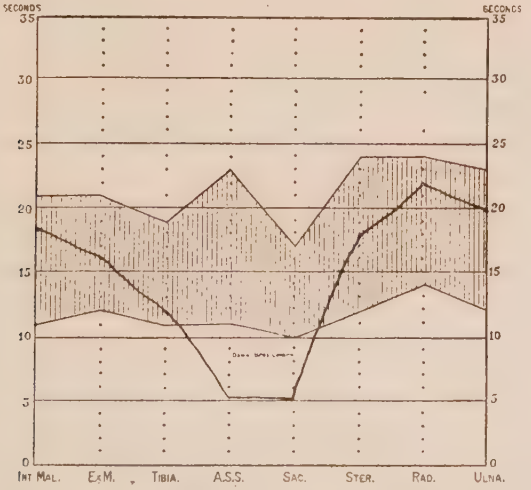

FIG. 4.—Tendon reflexes preserved. Marked analgesia of skin of face.

was plunged into the tip of his nose deep enough to hold it in position.

Vague gastric cases in which pain was the predominating factor studied by the vibration test proved the usefulness of the procedure. In some clinics gastric crisis as a very early symptom of tabes is frequently noted. Why there should be a difference in this observation in different places is not readily understood. In collecting the normal hundred on which the normal range was established gastric cases were sought after because of their non-neurologic character. This opened up a most interesting field, for it was found that in a number of instances these cases showed extraordinary abnormalities in the vibration response. In one case a tentative diagnosis of carcinoma had been made by a very capable house physician. Study of the nervous system showed gastric crisis in early tabes. In another case in the same ward ulcer had been diagnosed and the patient had been on rigid treatment for some time. Vibration study opened the way for a correction of the diagnosis, which was clearly gastric crisis in early tabes. Recently a case was studied in which the vibration response was distinctly that of tabes. Gastric pain was the symptom and a most careful chemical and fluoroscopic study had been made. The tendon reflexes were preserved and there was no Argyll-Robertson pupil, although the pupils were contracted. Analgesia of the skin of the face as well as other scattered areas suggested tabes, which was verified by spinal puncture. In still another case now under observation the diagnosis of carcinoma seemed well justified, but vibration suggested tabes, which was borne out by the Wassermann test and cell count of the spinal fluid.

In the eighty cases studied there has not occurred a single instance in which there was not a definite time reduction in vibration response which usually manifested itself in the form of the "sacral dip," in which other points of the lower half of the body frequently shared. It can be safely stated that any case with no reduction in the time of vibration response over the sacrum is not tabes—an observation which can be easily made in one-half a minute and relied upon without further investigation.

In multiple neuritis there was noted decided reduction in the

vibration time. The distinctive "sacral dip or notch," so marked in tabes, did not occur in any case. There was a general reduction of the time but a tendency for the sacral point to be relatively high and in many cases higher than the other points. In addition to this there was usually absent that tendency so marked in tabes for the time response of the upper extremity to be normal, making a curve which in that part of the body falls well within the normal range. In diabetes it was interesting to record the improvement in sensation indicated by the vibration curve in those cases receiving proper dietetic treatment. In a case of multiple neuritis occurring

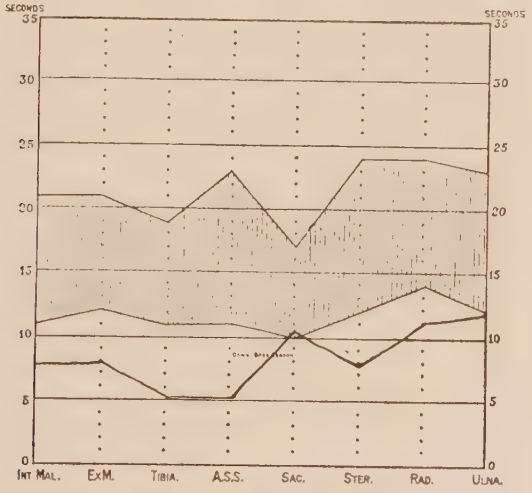

FIG. 5.—Multiple neuritis in pellagra.

in a woman who was a victim of pellagra seen in the service of Dr. Edwin Bramwell in the Royal Infirmary, Edinburgh, there was this definite reduction in vibration time with the high point over the sacrum. It was notable in this case that pain and tactile sensations were not disturbed and temperature sense only slightly. The vibration time is recorded in Fig. 5.

Three cases of Friedreich's ataxia were studied in Guy's Hospital and showed a loss in the vibration time quite like the changes noted in early tabes. The loss of vibratory appreciation was not complete and would have been overlooked if a qualitative test alone had been

made. As in tabes there is a "sacral notch" in the curve of these cases.

Subacute combined degeneration of the spinal cord in association with pernicious anemia was studied in five cases. In all cases there was a definite loss of the vibration response which, until late in the process, was confined to the lower half of the body, with little abnormality in the upper half. In one of these cases the loss was complete in the feet, marked over the tibiæ but not entirely lost, less over the anterior-superior spines and still less over the sacrum,

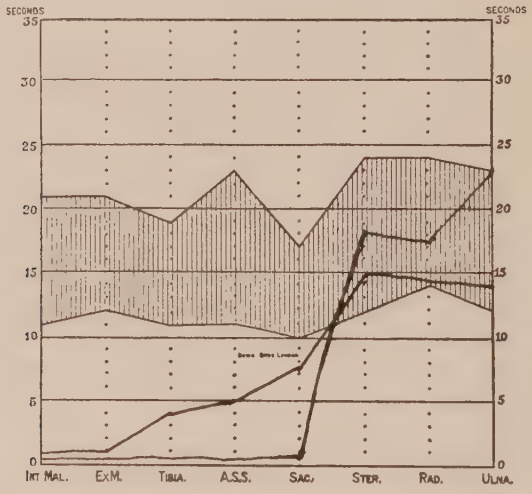

FIG. 6.—Two cases of subacute combined degeneration of the cord in pernicious anemia.

with a gradual ascent in the upper portion of the body. This is seen in Fig. 6, which is made of two such cases. In all of these cases the "sacral notch" of tabes was not pronounced. The curve can be readily distinguished from that of multiple neuritis.

One case studied caused much confusion because the response over the sacrum was entirely absent while the blood and spinal fluid were negative to the Wassermann reaction. Incontinence of bowel and bladder were present. Study of skin sensation about the anus and down the lower limbs gave the characteristic findings of a lesion of the cauda equina.

One of the most fertile fields for the study of the vibration sensation is syphilitic meningomyelitis. In this condition the vibration response was found to be reduced in the lower half of the body, but there was more evidence of asymmetry than is usually noted in such cord lesions as tabes. The great helpfulness of the test seems to be in gauging improvement. It seems probable that an improvement in the spinal fluid will run more or less parallel with an improvement in sensation, and this may be measured in a quantitative way by use of the vibration sensation. A beginning only has been made in this work, which requires much time and very exact laboratory work by an expert serologist.

In conclusion, I would thank Dr. A. F. Hurst, of Guy's Hospital, under whom the work was done, for aid of every sort; Dr. Gordon Holmes, for suggestions and clinical opportunities at the National Hospital in Queen Square, at Charing Cross Hospital and at Moorfields; Dr. Edwin Bramwell, for material and aid at the Royal Infirmary, Edinburgh.

SOME NOVEL EFFECTS PRODUCED BY STIMULATING THE NERVES OF THE LIVER

By W. B. CANNON, M.D., J. E. URIDIL, M.D.
AND
F. R. GRIFFITH, M.D.
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(ABSTRACT)

After removal of the adrenal glands, splanchnic stimulation will cause a faster beat of the denervated heart. Since this occurs when the inferior cava and the portal vein are closed below the liver, and since it does not occur when the hepatic nerves are cut, the inference is drawn that some agent arising in the liver is the occasion of the faster beat.

Stimulation of the hepatic nerves will cause an increased rate of the denervated heart, an effect appearing later than the similar adrenin effect and lasting for a longer time.

Stimulation of the hepatic nerves will cause a rise of blood-pressure. Evidence is adduced to show that this is not the consequence of a retarded blood flow through the liver; it does not occur on closure of the hepatic artery and vein. It occurs on hepatic stimulation, although all abdominal viscera have been removed except the liver, and unlike stimulation of splanchnic bloodvessels alone it long outlasts the period of stimulation.

Asphyxia is very promptly depressive in its influence on the effectiveness of hepatic stimulation.

The increments of heart-rate vary widely. They are slight if the animal has been fasting or is in poor condition; they are much greater if the animal is digesting meat. The inference is drawn that

a substance is given off by the liver into the blood stream which, carried to the denervated heart, raises its rate.

None of the known or supposed products of hepatic activity—glucose, urea, catalase—when injected into the blood stream have the effects produced by exciting the hepatic nerves.

Watery extracts of the liver are ineffective. Liver extracted by boiling acid and nearly neutralized augments the heart-rate, but so do extracts of other organs.

The efficacy of hepatic stimulation in causing a faster heart-rate when meat is being digested is not seen if an animal is digesting carbohydrate or fat or has been fed for several days on either of these foodstuffs. On the other hand, stimulation is more effective after amino-acids have been injected into the intestines.

The tentative conclusion is drawn that probably the effects noted are not due to a true internal secretion produced by the liver but to a discharge from its cells of amino-acids or amines, which are sympathomimetic in character.

DISCUSSION

Dr. H. Sewall, Denver, Col.: Did the doctor make any observations on the volume of blood entering the heart? Was it increased? That would explain the increased pulse-rate on other grounds than those which he has mentioned.

Dr. Walter B. Cannon (closing): We have gone carefully through the literature and have failed to find any record of variations in the rate of the denervated heart due solely to changes in arterial pressure or in the rate of flow through the coronary arteries. This testimony is thoroughly supported by numerous observations of our own.

NEW EVIDENCE OF THYROID SECRETION FOLLOWING STIMULATION OF THE CERVICAL SYMPATHETIC

By W. B. CANNON, M.D.
AND
P. E. SMITH, M.D.
BOSTON, MASS.

(ABSTRACT)

Gentle massage of the thyroid gland in the cat for two or three minutes will cause an increased rate of the denervated heart, amounting in some instances to 33 per cent over the basal rate. The development of the maximal increase of rate is usually slow, requiring from thirty to sixty minutes, and passing off in a similarly slow manner.

Massage of another gland, *e. g.*, the submaxillary, does not cause this effect.

The augmentation of heart-rate caused by thyroid massage occurs in the absence of the adrenal glands.

Stimulation of the cervical sympathetic trunk as it leaves the stellate ganglion induces a similar augmentation of the rate of the denervated heart; this does not occur if the thyroid gland has previously been removed.

If the cardiac fibers from the stellate ganglia are severed, as well as the vagus nerves, and an afferent nerve, such as the sciatic or brachial, is stimulated under a degree of anesthesia which will permit reflex retraction of the nictitating membrane and dilatation of the pupil, there is a primary increase of rate due to adrenal secretion, followed by the slowly developing increase characteristic of the thyroid effect.

If the vagi and the cardiac fibers of the stellate are cut and the animal is asphyxiated under conditions which permit the eye changes described above, there is a similar primary rise due to adrenal secretion, followed by the secondary thyroid effect.

If the thyroid glands have been previously removed, sensory stimulation and asphyxia induce only the increase of rate due to adrenal discharge.

INFLUENCE OF INORGANIC IRON ON THE REGENERATION OF THE BLOOD AFTER ANEMIA

By JOHN H. MUSSER, Jr., M.D.
PHILADELPHIA, PA.

(ABSTRACT)

Last year Dr. Whipple and his co-workers presented a series of papers which were most stimulating and yet were rather radical in view of our present-day belief in the efficacy of iron in the treatment of anemia. These authors held that in the treatment of ordinary anemia inorganic iron has little if any effect upon the regeneration of blood without adequate diet, and they held that "It is impossible to assume without positive proof that inorganic iron is of value in the treatment of simple anemia." The long controversy that has existed since 1746, and which was apparently settled in 1906, might be said to have been reopened by these series of papers.

The present studies were undertaken in order to test out or to confirm these apparently revolutionary ideas in animals that had been repeatedly deprived of small amounts of blood. Anemia of this type undoubtedly resembles very closely the type of anemia, which we see clinically, secondary to the loss of small amounts of blood.

Methods. The animals were bled at varying intervals and a test animal was put upon iron adequately controlled. Following the administration of the iron the following studies were undertaken: Hemoglobin estimation; red and white cell counts; differential count; resistance of the red cells; percentage of skeined cells; determination of the total blood volume. The animals were

given iron in the form of equal parts of ferrous sulphate and sodium bicarbonate, and a dose was given which was equivalent to that of 1 gram a day to an individual of 65 kilos.

Experimental Data. In the first experiment two splenectomized dogs were employed which had been rendered anemic as a result of weekly bleedings of 100 cc for two and a half months. These dogs were observed for a period of nearly two months; blood counts were made every second or third day. In this experiment we may say briefly that the dog that was given the iron returned more rapidly toward normal figures than did the control dog. However, the control animal started with a lower figure than the test animal, and both animals obtained the highest figures at about the same time.

In the second experiment dogs were used that had not been splenectomized. The experiment was somewhat similar to the first. In this experiment the two animals approached normal at approximately the same time, but the return of the control animal to normal was more rapid and more complete.

In the third experiment two dogs which had been bled over a short period of time were bled large quantities for six consecutive days, and were then followed as in the other experiments. In this experiment the control dog apparently regenerated more rapidly than did the test animal.

In the last experiment, two animals were selected which had been splenectomized and then rendered anemic by hemorrhage repeated on two successive days. In these two animals it can be said that both returned to normal in about the same length of time, and the iron had practically little effect on them.

Discussion. The results of these experiments would seem to substantiate the contention of Whipple that under ordinary circumstances the giving of inorganic iron does not have a particularly marked effect upon blood regeneration and the correction of anemia in experimental dogs. It may be contended that the return to the normal in these animals is so rapid that the iron can have but little effect when Nature is taking care of the anemia so thoroughly. But in two of the dogs, which had been rendered anemic by repeated bleedings and by a long splenectomy, the return to normal was

relatively slow, yet in neither the control, 19–6, nor the experimental dog, 19–4, was there any obvious or material difference in the rapidity of the regeneration of the red cells or hemoglobin. Differences, of course, exist between the dog taking iron and the controls, but only such as would be expected. It would be almost impossible to obtain animals which were alike in every respect. The hemopoietic systems of no two animals will respond in exactly the same manner. In considering experimental work of this type various factors must be taken into consideration. The resistance of the red cells to hypotonic salt solution in some animals is greater than in others, and this must play some factor in altering the course of the anemia. In some animals the response of the marrow and the stimulation of hemorrhage is greater than in other animals. This can be demonstrated by the skein cells. In some animals the blood volume is greater and better sustained than in others. Lastly, there is no absolute mean normal count, as these various factors produce differences in the hemoglobin and red cell counts in the different dogs.

SUMMARY. In two of four experiments dogs receiving iron regenerated blood more rapidly than did their controls. In the other two experiments the control dogs apparently improved more quickly than did the test dogs, and this improvement is found to be particularly definite when analyzed in relation to the blood volume changes.

CONCLUSION. The administration of inorganic iron cannot be said to produce any constant alteration in the course of an experimental hemorrhagic anemia.

APPLICATIONS OF SALIGENIN AND OTHER AROMATIC ALCOHOLS AND THEIR DERIVATIVES TO THE PROBLEMS OF THERAPEUTICS

By ARTHUR D. HIRSCHFELDER, M.D.
MINNEAPOLIS, MINN.

Most of our medicines have come to us ready-made in plants and have been handed down to us by the savages or folk-lore of old ladies. And the real patrician substances of our materia medica, the alkaloids, are of very humble origin, for they are merely the unexcreted waste products in the metabolism of plants. Our medical forefathers have not been above introducing the excreted waste products of human metabolism into the rich materia medica of bygone days; but though we have advanced from this era we are still in the stage of a general feeling that the substances produced by the leaves of brainless plants are better as medicines than the substances which can be created in the laboratory for definite pharmacologic purposes by the brain and hand of the chemist.

Just a glance at the tremendous structures of the alkaloid molecules like morphin, strychnin and cocain show that they are so big and complex that only a small portion of their structure can be the one actually useful in theory, and the rest is either useless or adds to their toxicity. This was established by Stahlschmidt in 1859, when he showed that the mere addition of methyliodide to the strychnin molecule changed its action from the well-known convulsant effect to a paralyzing action like that of curare.

The medicine of the future must deal with simpler molecules, stripped of all the unnecessary toxic portions down to the smallest and simplest molecules that will do the work of therapeutics.

It was therefore a great step in advance when David Macht[1] showed that the local anesthetic action of papaverin was due solely to the benzyl group and could be simulated by benzyl alcohol, and that its antispasmodic action could be simulated by benzyl benzoate.

It seemed of interest to me to determine whether of such simple substances benzyl alcohol was the one best adapted to local anesthesia or whether other similar substances of the same series were as good or better local anesthetics. Accordingly, two years ago[2] we investigated a number of other members of the series, phenylethyl alcohol,[3] cinnamic alcohol, piperonyl alcohol, phenylglycol, saligenin, with regard to their local anesthetic properties and their toxicity. We found that saligenin, the alcohol corresponding to salicylic acid, is distinctly the best local anesthetic of this series. It has about the same toxicity as benzyl alcohol, but it keeps much better, is much more certain in its anesthetic properties and is much less irritant to the subcutaneous tissue. It is five to ten times less toxic than procain and twenty-five to fifty times less toxic than cocain. In fact in an intravenous injection in mammals the lethal dose is 0.4 to 0.7 gram per kilo, while the subcutaneous injection of 1 gram per kilo causes only a transitory paresis in rabbits and is not fatal. This would correspond to 70 grams of the solid substance or 3 liters of a 2 per cent solution subcutaneously, a hundred times as much as is necessary for the ordinary minor operation.

Accordingly, this substance has been tested by a number of surgeons in Minneapolis and elsewhere and has been found to give infiltration anesthesia, which has been uniformly satisfactory for minor operations, such as tonsillectomies and the removal of small tumors, and also for a number of major operations, such as thyroidectomies, herniotomies and laparotomies.

Saligenin resembles novocain rather than cocain in that it does not anesthetize the mucous membrane of the mouth, although it does anesthetize the conjunctiva and to a great degree the urethral mucosa.

[1] Jour. Pharmacol. and Exper. Therap., 1918, xi, 263.

[2] Hirschfelder, A. D., Lundholm, A., and Norrgard, H.: Jour. Pharmacol. and Exper. Med., 1920, xv, 261; Science, p. 19.

[3] This substance was also investigated independently by Hjort and Eagan, Jour. Pharmacol. and Exper. Med., 1919, xiv, 211.

Dr. H. M. Wynne[1] has obtained anesthesia satisfactory for cystoscopy in women by the use of 4 per cent solutions of saligenin intra-urethrally in a series of 26 cases; and in men Dr. Wethal has found similar results in 8 cases and Dr. Thomas, using 8 per cent solutions, has had the same experience in 20 cases, even in some with very tender urethras.

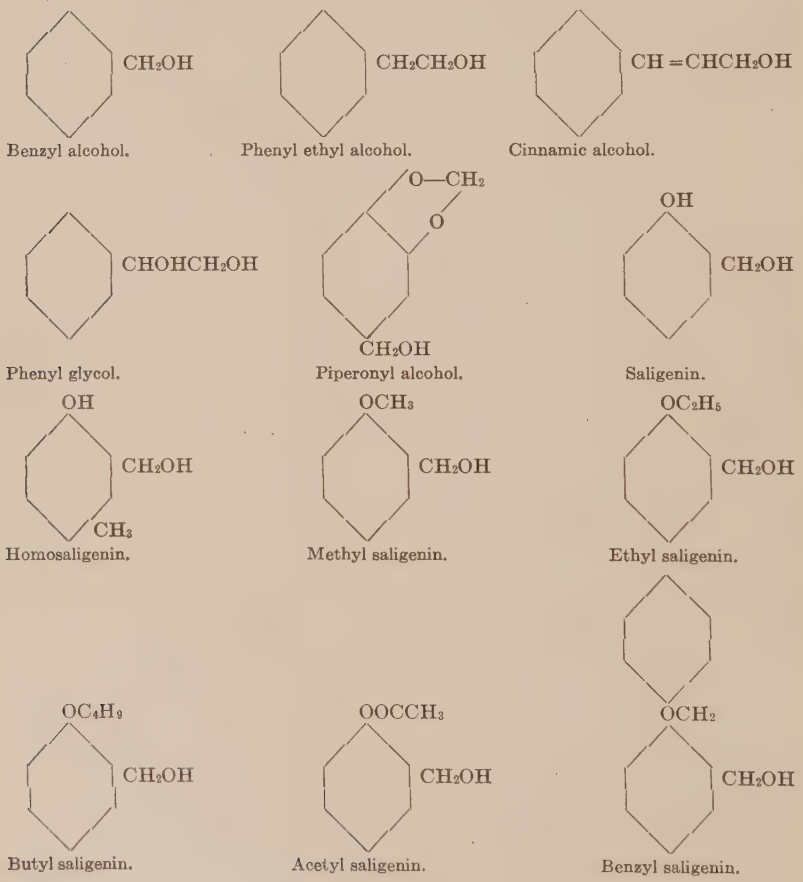
Ointments of saligenin in petrolatum have proved very useful for the relief of a number of conditions. In 7 cases of nursing women with painful fissured nipples, under the care of Drs. W. R. Shannon and E. J. Huenekens, applications of 2 per cent ointments enabled nursing to take place painlessly and seemed to assist in the healing; but in a seventh case Dr. Huenekens reports that it caused some irritation. In several cases of large burns saligenin ointment caused marked relief from pain, though on account of the salicyl content its use should not be continued for more than a day or so. Dr. Shannon reports good effects of the ointments for the relief of itching in very annoying cases of eczema in children, though here again the use must be confined to short periods. They have also proved useful for the treatment of stubborn pruritus ani and for the itching along the lineæ albicantes in a pregnant woman. Five per cent ointments applied to the skin produce definite lowering of sensitiveness, though not absolute anesthesia. When applied to the mucous membrane of the prepuce this gives sufficient anesthesia to prevent all painful sensation in the treatment of venereal warts. Dr. A. Sweeney, of St. Paul, Minn., has found that the use of a 5 per cent ointment of saligenin made it possible for a man who had a continuous catheter inserted through a fistula of the abdominal wall after a suprapubic cystotomy to keep the catheter in and enabled it to be inserted and removed without pain, to the great relief of the patient.

Saligenin also has a considerable analgesic action when administered by mouth. This was tested by administering the drug in $\frac{1}{2}$-gram capsules three times per day for two-day periods, alternating with periods during which the patients received sodium salicylate or acetyl salicylic acid. In 2 cases of acute arthritis and in 4 cases

[1] Jour. Am. Med. Assn., 1920, lxxv, 1770.

of chronic arthritis, under the care of Dr. H. Lippmann and Dr. H. Rypins, it was found that ½-gram doses were about equal in therapeutic effects to 1-gram doses of salicylates. In none of these cases were untoward symptoms or disturbances of the kidney encountered, though one medical student with chronic arthritis became nauseated from the use of ½-gram doses of saligenin.

FIG. 1.—STRUCTURAL FORMULÆ OF SOME PHENYL CARBINOLS.



We had hoped that by alterations in the molecule we might obtain substances with a true cocain effect, and we therefore investigated the effects of various alterations in the molecule.

J. P. Quigley,[1] one of our graduate students, has found that if some other radical is substituted for one of the hydrogens in the CH_2OH group of benzyl alcohol and anesthetic, action is lessened, and if both are substituted for the anesthetic, action is lost. On the other hand ethers of saligenin even in 1 per cent emulsions produce definite numbing of the tongue and give the same peculiar after-taste and numbing sensation which is usually regarded as

FIG. 2.—TABLE SHOWING RESULTS IN A SERIES OF CYSTOSCOPIES DONE BY DRS. G. P. THOMAS AND MELLIN, USING SALIGENIN (8 PER CENT) AS THE ANESTHETIC.

Name.	Age.	Condition.	Procedure.	Result of anesthetic.
A. S. B.	53	Enlarged prostate	Cystoscopy	Very good.
J. T.	19	Cystitis	Cystoscopy	Very good.
F. J.	54	Kidney stone	Cystoscopy	Very good.
M. H.	45	Negative	Cystoscopy	Very good.
A. J.	57	Bilateral introvesicular enlarged prostate	Cystoscopy	Good, slight edema at meatus.
K. D.	20	Hydronephrosis	Cystoscopy	Good.
J. T.	19	Cystitis	Cystoscopy	Good.
M. C.	33	Slight prostate	Cystoscopy and cautery	Good.
T. A. S.	56	Bilateral introvesicular prostate	Cystoscopy	Fair.
M. K.	56	Carcinoma bladder	Cystoscopy	Very good.
Dr. B.	30	Stricture	Urethroscopy	Very good.
M. P.	52	Papilloma bladder	Cystoscopy	Very good.
M. C.	57	Papilloma bladder	Cystoscopy	Very good.
J. P.	22	Cystitis pyelonephritis	Cystoscopy	Very good.
M. B.	52	Negative	Cystoscopy	Fair only.
M. G.	62	Tuberculosis of chest and bladder	Cystoscopy	Good.
M. R.	30	Negative	Cystoscopy	Poor (lues?)
M. S.	34	Negative	Cystoscopy	Fair.
J. C.	34	Pyelitis	Cystoscopy	Very good.
F. H.	41	Bladder stone	Cystoscopy and lithopaxy	Very good.

characteristic of cocain. However, they also give a burning sensation which precludes their practical use upon mucous membranes, and on subcutaneous injection the methyl and ethyl ethers cause definite edema and irritation.

The ethers of saligenin cause fall of blood-pressure and also inhibit the contraction of smooth muscle just as do the benzyl esters,

[1] Jour. Pharmacol. and Exper. Therap., 1921, xvii, 326.

(Fig. 3), though I am not yet certain whether their effect is any greater. The benzyl ether seems to have the most lasting effect in lowering blood-pressure.

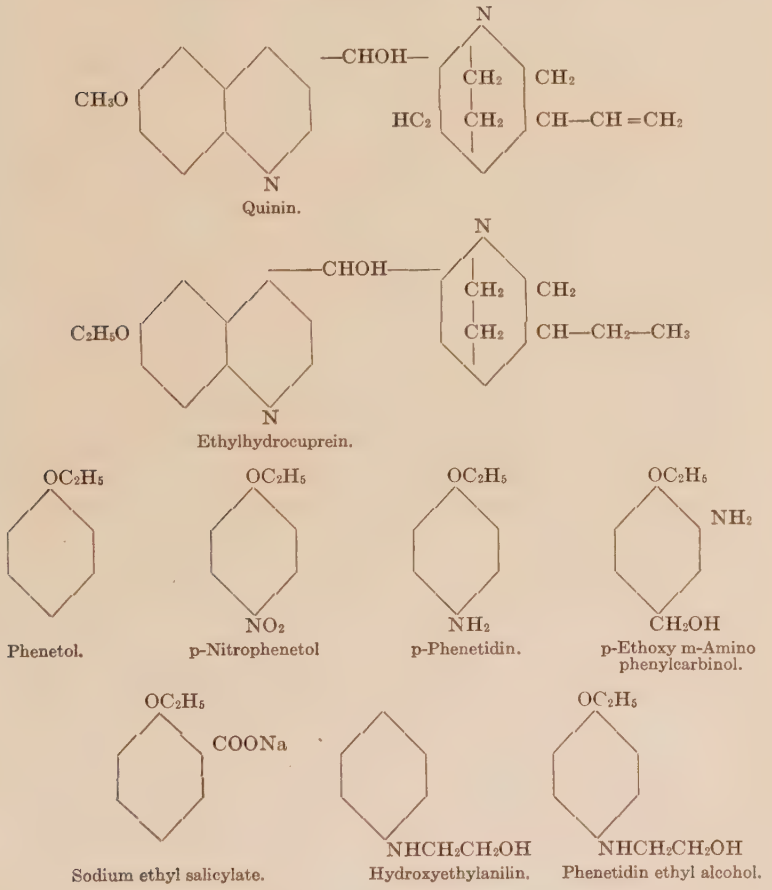
FIG. 3.—Tracing showing the effect of the benzyl ether of saligenin upon the contractions of an isolated loop of rabbit's small intestine. 0.5 cc. 2 per cent benzyl ether of saligenin introduced into the Ringer-Locke solution at ↑.

These substances, as you will notice, are all nitrogen-free compounds, which, like the substances described by Dr. Macht, can do the work of alkaloids.

It was of considerable interest to determine whether the mere presence of nitrogen in a similar compound would be of real advantage to its action. Accordingly, we have tested a number of substances, such as hydroxyethylanilin and parethoxy meta amino

phenyl carbinol, the substance whose structure is shown on the screen. Both of these substances contain a benzene ring, a basic nitrogen group and an alcohol group. Hydroxyethylanilin in 2 per cent solution is very slightly anesthetic on subcutaneous injection in man; the other substance is not at all so, though both exert an anesthetic effect upon the skin and the sciatic nerve of a frog.

FIG. 4.—STRUCTURAL FORMULÆ OF QUININ, ETHYLHYDROCUPREIN AND SOME ETHOXY COMPOUNDS.



It is evident, therefore, that the mere presence of nitrogen in the molecule does not contribute much to its pharmacological action

provided the other groups are active ones. The best substances of the series we have thus far studied are those which combine a low but definite water solubility with a high degree of solubility in organic solvents and in lipoids.

One thing that has been striking is that of all the substances that we have thus far investigated, all those which are active physiologically, have a somewhat bitter taste. It seems to me, therefore, that it is more than a coincidence that both little children and the doctors find that most good medicines have bitter tastes. It is probably because most medicines owe their efficiency to the fact that they act on the nerves—and the nerves of taste, and particularly those that distinguish bitter tastes, are the first of all the nerves to respond.

We have also been interested in these simpler substances from another angle, namely, the possible relation of chemical structure to their antiseptic and chemotherapeutic action, particularly upon the pneumococcus, the gonococcus, the trypanosome and the spirilla. Mortgenroth, in 1911, demonstrated that if an ethyl group is substituted for a methyl group in quinin or hydroquinin the substance develops a strong pneumococcidal and trypanocidal action both *in vitro* and *in vivo*. The ethyl hydrocuprein molecule and some of our anesthetic substances have two points in common—a benzene ring structure and an alcohol (CHOH) group. With the ethyl saligenin it also has an ethoxy group in common. We have attempted to determine whether other aromatic ethoxy compounds, with or without the carbinol group, would have pneumococcidal and trypanocidal actions, for if so this might furnish a new starting-point for the development of more active chemotherapeutic substances.

We have therefore studied the effects of a considerable number of aromatic compounds, phenetol, nitrophenetol, phenetidin, phenetidin ethyl alcohol, ethyl sodium salicylate and para ethoxy-meta aminophenyl carbinol, comparing their antiseptic and bactericidal action *in vitro* with that of corresponding compounds which did not contain the ethoxy group, but we have been unable thus far to find any evidence of specific action in these simpler ethoxy compounds. Most of the aromatic alcohols have a mild antiseptic action but present no striking features in this regard.

None of the compounds which we have studied thus far kill the Trypanosoma brucei or the Spirillum obermeyeri in rats.

Since we have been unable to determine any relation between the presence of the ethoxy group and any specific action upon pneumococcus and trypanosomes, we are also comparing the physicochemical properties of quinin and ethyl hydrocuprein, such as relative solubility in water and in ether and oil and their effect on surface tension of water, and at the interface between oil and water and between water and oil containing kephalin, and these will be reported on at a later date.

EXPERIMENTAL INQUIRY INTO THE CEREBRAL AND NEUROMUSCULAR MANIFESTATIONS OF DIGITALIS BODIES

By D. I. MACHT, M.D.
AND
WILLIAM BLOOM, A.B.
BALTIMORE, MD.

Cerebral symptoms in the course of heart disease are not at all uncommon. Everyone is familiar with the classic work of Head[1] and several other interesting publications on the subject, such as by Riesman[2] and others. There is, however, one group of psychoses occurring in cardiac patients which has received perhaps insufficient attention on the part of clinicians. These are the patients exhibiting hallucinations, delirium and other mental affections occurring at the height of digitalis therapy. The father of rational digitalis therapeutics, Withering,[3] did not fail to note certain cerebral manifestations of digitalis, and mentions them casually in his famous study of the foxglove. The most important contribution on this subject, however, came from an eminent French physician, Duroziez,[4] in 1874. He gives a list of twenty cases in which delirium or hallucinations with or without death accompanied the administration of digitalis, and as he believed were caused thereby. Dr. H. O. Hall,[5] in 1901, revived the interest in the subject by an article published in *American Medicine*, in which he reviewed the cases of Duroziez and others and described one from his own experience.

[1] Brain, 1901, xxiv, 345.
[2] Tr. Assn. Am. Phys., 1920, p. 77.
[3] An Account of the Foxglove, Birmingham, 1785.
[4] Gaz. hebd. de méd. et de chir., 1874, ii, 780.
[5] Am. Med., 1901, i, 598.

We have become interested in the question whether the digitaloids produce any cerebral effects in connection with the study of the action or influence of various drugs on the behavior of white rats in the circular maze,[1 2 3] and we have performed a series of experiments with various digitalis bodies in order to clear up, at least partially, this subject. The method of studying the behavior of rats in the circular maze is briefly as follows: Young adult and perfectly healthy albino rats are trained to find their way from the periphery of a circular maze to the center in the shortest period of time without committing any errors. The maze consists of a series of concentric circular alleys connected at irregular intervals by passages and provided with an equal number of blind cul de sacs. The animal is taught to meander its way correctly through the labyrinth to the center of the maze, where some food is placed. Healthy rats of good pedigree can learn to solve the maze problem in a period of from ten to eighteen days. They will then run immediately from the peripheral entrance of the maze to the center by the shortest route without running into any of the blind alleys. The running time for such a performance may be as brief as four seconds. The effect of various drugs, such as narcotics, antipyretics, etc., has been studied by the authors by determining first the normal running time of a rat, then injecting with the drug in question and after suitable periods of time observing the behavior of the animal in the maze again. An analysis of the data furnished by the maze has been described elsewhere. By means of this apparatus we are enabled to study not only the learning time and the "memory habit" of the animals, but also their neuromuscular coördination and general behavior.

In the present research the following drugs were injected into rats and their effects on behavior studied: Ouabain; amorphous strophanthin; digitoxin; digitonin; various samples of digitalin; digalen; several injectable preparations of digitalis leaf, such as digitotal, digipuratum and digifolin; and lastly the curious heart drug, closely

[1] Macht, D. I., and C. F. Mora: Jour. Pharmacol. and Exper. Therap., 1920, xvi, 219.

[2] Macht, D. I., and Bloom, Wm.: Ibid., 1921, xvii, 21.

[3] Macht, D. I., and Bloom, Wm.: Arch. internat. de pharm. et de therap., 1920, xxv, 8.

related to the digitaloids, discovered by Abel and Macht, and named bufagin.

A total of thirty rats were used in this investigation. It was found that all of the above substances were very depressant to the rats, markedly affecting their behavior, memory habit, discrimination and neuromuscular coördination even when injected in not very large doses. This was especially interesting in view of the fact that rats are notably resistant to the digitaloid poisons, so that the results obtained were not merely the effects of lethal doses of the drug but were more or less comparable to the effects that would be produced in patients by full therapeutic doses.

It would seem, therefore, that digitaloid bodies do exert a depressant effect on the cerebral and neuromuscular functions. The authors have sent letters of inquiry to a number of prominent clinicians asking whether they have noted any cases of cerebral manifestations which could be correlated with the administration of full doses of digitaloids. So far only one of them stated that he had seen a few such cases which seemed to be directly referable to digitalis as the causative agent. It is hoped that the present article may revive interest in the subject and that more clinical data may be reported concerning the cerebral effects of digitaloid drugs.

Fuller data will be published in the *Archives of Internal Medicine*.

THE ACTION OF BACTERIAL CULTURE PRODUCTS ON PHAGOCYTOSIS[1]

By AUGUSTUS B. WADSWORTH, M.D.

AND

E. N. HOPPE

Few of the bacterial poisons have been isolated or identified by chemical analysis; the true toxins are scarcely known save for the reactions they induce in animals. They can be recognized and their potency measured only when these reactions are characteristic as they are with tetanus, diphtheria and botulinus toxins. The febrile reactions and the development of immunity that are induced by other pathogenic species of bacteria in the course of many of the diseases of man suggest quite definitely the production of toxic substances capable of inciting specific immune reactions, but it has been impossible to detect or to measure accurately these substances.

This study was undertaken not only for the purpose of investigating further the action of the known bacterial poisons on the body cells, but also in the hope of finding some isolated animal cells or tissues sensitive to the unidentified bacterial substances and a technic delicate enough to register degrees of injury to their normal reactions.

The first cells selected for this investigation, and the only ones dealt with as yet, were the leukocytes. These simple cells were chosen for the obvious reason that they were eminently suited for a study involving elementary life phenomena. They could be easily isolated; they would react to a variety of alterations of their

[1] From the Division of Laboratories and Research, New York State Department of Health, Albany, N. Y.

environment and those alterations could be quantitatively and qualitatively controlled.

Technic. A very simple technic was evolved after a long series of preliminary studies. Each operation was standardized. In this technic the phagocytic activity of the leukocyte for sensitized staphylococcus cells was selected as the index by which to evaluate the degree of the action of the test agents. In general the method consisted of exposing well-washed leukocytes to the action of the substances to be tested. The effect of this exposure was then measured by the degree of phagocytosis that occurred when sensitized staphylococci were brought in contact with the leukocytes. This degree was determined from microscopic preparations and stated in units as the phagocytic index.

The principle observed in developing the technic was that only one unknown factor should be present in each test and the elements of this unknown factor carefully controlled. The difficulties encountered in this process are exceedingly well pointed out and discussed by Fleming in 1908.[1] The details of the finished technic are given here:

The materials used in the tests were: (1) A suspension of washed leukocytes; (2) an even emulsion of sensitized *Staphylococcus pyogenes aureus,* and (3) various toxins and culture broths which are collectively called test substances.

The leukocytes were obtained from the circulating blood of a normal dog by bleeding it with a syringe from the jugular vein. For each 1 cc. of leukocyte suspension required, 10 cc. of blood were taken. This blood was carefully mixed with an equal volume of 2 per cent sodium citrate solution to which 0.5 per cent sodium chloride had been added. It was immediately distributed in 15 cc. pointed centrifuge tubes, 10 cc. in each, where the white blood cells were collected in strata by centrifugalizing for thirty minutes at low speed, No. 3 on the rheostat (size 1, type B centrifuge, manufactured by the International Equipment Company). High-speed centrifugalization mutilated the

[1] Some Observations on the Opsonic Index with Special Reference to the Accuracy of the Method and to Some of the Sources of Error, Practitioner, 1908, lxxx, 607–634.

leukocytes. The leukocyte strata were removed from the tubes by the usual capillary pipette method and washed in ten times their volume of 0.85 per cent salt solution. The leukocytes were again separated by centrifugalizing and drawn off as before. The final suspension was then made in 0.85 per cent salt solution in the proportion stated above, *i. e.*, each 10 cc. of blood to contribute 1 cc. of leukocyte suspension. Many red cells were retained, but this was no disadvantage. Leukocytes obtained by aleuronat injections into the peritoneal cavity of small laboratory animals were quite unsuited because of their lack of uniformity in phagocyting after being wholly or partially filled with aleuronat. Leukocytes from rabbits' blood were also rejected because of their extreme variation in number per cc. in different rabbits. There were no objections to leukocytes from horses' blood, but these were not as readily obtainable as dog leukocytes.

The staphylococci were grown eighteen hours on a beef-infusion agar slant which was inoculated from a seed culture transferred every two or three days. The organisms from this eighteen-hour culture were suspended in 8 cc. of 0.5 per cent salt solution and sensitized by adding 0.1 cc. of normal horse serum. This mixture was thoroughly agitated by repeatedly drawing it into and forcing it from a capillary pipette. It was then incubated at 37° C. for fifteen minutes. The organisms were quickly thrown down by centrifugalization. The supernatant fluid was discarded and replaced by 10 cc. of 0.5 per cent salt solution. The organisms were thoroughly separated and washed in this solution with a capillary pipette. After centrifugalization this washing was discarded and the staphylococci were evenly suspended in 0.5 per cent salt solution to give an opacity equal to that of a No. 8 barium sulphate scale. A perfectly homogeneous suspension of the staphylococcus was more easily maintained in 0.5 per cent than in 0.85 per cent salt solution. Sensitization with normal horse serum was preferable to sensitization with staphylococcus immune rabbit serum because the organisms were more slowly and uniformly ingested, and beyond a certain point numbers of organisms in a leukocyte were not desirable. The number of staphylococci in the final suspension was given special attention—as was the case with the final leukocyte suspension—for at best this varied con-

siderably. Controls were always included, but uniformity of results in the separate tests was desired in so far as it could be maintained and the ratio of staphylococci to leukocytes was a factor.

The action of the culture material of different species under different conditions was tested with the leukocytes and staphylococci and constituted the variable factors in the experiments. The descriptive details of these are more logically and conveniently considered in connection with the tests in which they were used and are described in that connection.

The tests were performed *in vitro*.

Small test-tubes, 1 cm. by 7 cm., were set up in racks and labeled. A tube was provided for each test substance and an additional tube for each factor that required a control. Then 0.1 cc. of each test substance or control substance was pipetted into its allotted tube and 0.2 cc. of the leukocyte suspension was mixed with each. The racks of tubes were incubated at 37° C. for fifteen or thirty minutes for the test substances to act on the leukocytes. After this exposure 0.1 cc. of the sensitized staphylococcus suspension was added to each tube and the contents were mixed as before. The racks were returned to the incubator and twenty-five minutes were allowed for phagocytosis to take place.

Smears were made from the phagocytic mixtures upon glass slides in the ordinary manner of blood-smearing and stained in Coplin jars with Wright's stain.

At least fifty polymorphonuclear leukocytes were counted and the following indexes determined: (1) The percentage of leukocytes phagocyting, (2) the average number of staphylococci ingested by the phagocytes, and (3) the phagocytic index, which was the average number of staphylococci ingested by the fifty leukocytes or by as many leukocytes as were counted.

Phagocytic technic *in vitro* has generally fallen into disuse on account of the extreme difficulty in securing reliable results with it. The numerous investigations and discussions of the purely technical phases of the problem will be omitted here. However, in working out the present technic it was obvious that variations in almost any factor could materially affect the final results. Even faulty technic in smearing a single slide could mutilate enough phagocytes to

vitiate the results of an entire test. The possibilities for error were so numerous that none of this work was entrusted to assistants. Results were discarded that could not be readily duplicated. No conclusions were drawn unless the results of the experiment showed a definite and well-marked effect on phagocytosis.

Experiments. In the first series of experiments the action of a variety of toxins and culture broths was tested on the leukocytes.

Standard diphtheria toxin was selected for the preliminary study, and it was found that after leukocytes had been exposed to its action for fifteen minutes they ingested almost no sensitized staphylococci as compared with the control preparations.

Then fifteen other toxins and culture broths of widely differing bacterial species were tested. The results were similar in every case to those obtained with diphtheria toxin. The extent of the inhibition of phagocytosis is well shown in the photomicrographs reproduced at the end of the paper.[1]

In the table the toxins and culture broths and the controls are listed and described in the second column. The headings of the succeeding vertical columns follow the order of the steps in the established technic and the last three columns give the resulting indexes.

From the phagocytic indexes of this table it is readily seen that the potency of the toxin as indicated by the minimal lethal dose did not affect phagocytosis.[2] Toxin No. 2 with a minimal lethal dose of at least 10 cc. was produced by an attenuated culture of the same strain that produced toxin No. 1 with a minimal lethal dose of 0.003 cc. The culture medium was similar in both cases. Yet both toxins were equally depressing to the phagocytic index. Also it may be seen that the hydrogen ion content was not a factor here; for example, the pH value of toxin No. 1 was identical with that of its control broth.

[1] This paper with the illustrations appears in the Journal of Immunology, 1921, vi, 399.

[2] It is significant here to note the results of another study of the action of the bacterial toxins on the leukocyte (Wadsworth, A. B., and Vories, R.: The Action of Leukocytes and Brain Tissue on Diphtheria and Tetanus Toxins, Jour. Immunol., 1921, vi, 413). In mixtures of diphtheria and tetanus toxin with leukocytes no loss in the potency of the toxin could be detected by either the minimal lethal dose or the intracutaneous test. Apparently there is no physical or chemical combination between these true toxins and the leukocytes under the conditions of the experiment.

THE ACTION OF TOXINS AND CULTURE BROTHS IN DEPRESSING THE PHAGOCYTIC POWER OF LEUKOCYTES

	Toxins, culture broths and controls.	Cc.	Dog leukocyte suspension, cc.	Exposure of leukocytes to toxins and broths at 37° C., min.	Sensitized staphylococcus, cc.	Incubation at 37° C., min.	Average number of bacteria in a phagocyte.	Percentage of phagocyting leukocytes.	Phagocytic index.
1	Diphtheria toxin No. 161,[1] M. L. D. 0.003 cc., 7-day culture, pH 7.4	0.1	0.2	15	0.1	25	8.50	20	1.70
	Control diphtheria toxin broth, pH 7.3	0.1	0.2	15	0.1	25	26.13	88	23.00
2	Diphtheria toxin No. Um_2[2], M. L. D. 10 cc. +, 7-day culture	0.1	0.2	15	0.1	25	5.40	20	1.08
	Control diphtheria toxin broth, pH 7.3	0.1	0.2	15	0.1	25	23.14	70	16.20
3	Tetanus toxin No. 93,[3] M. L. D. 0.0001 cc., 14-day culture	0.1	0.2	15	0.1	25	7.13	30	2.14
	Control tetanus toxin broth, pH 6.7	0.1	0.2	15	0.1	25	23.40	94	22.00
4	Botulinus toxin No. 1B, M. L. D. 0.005 cc., 10-day culture[3] pH 6.2.	0.1	0.2	15	0.1	25	9.34	46	4.30
	Control Van Ermengen broth, pH 7.5	0.1	0.2	15	0.1	25	28.55	76	21.70
5	Tuberculosis culture broth, virulent human strain No. 160, 21-day culture	0.1	0.2	15	0.1	25	14.23	26	3.70
	Control glycerin broth, pH 7.4	0.1	0.2	15	0.1	25	23.04	92	21.20

6	B. prodigiosus culture broth, 18-day culture, pH 7.8	0.1	0.2	30	0.1	25	12.14	14	1.70
7	Streptococcus culture broth, 18-day culture, pH 6.2−	0.1	0.2	30	0.1	25	10.50	28	2.94
8	Pneumococcus culture broth, virulent strain, 10-day culture[4]	0.1	0.2	30	0.1	25	20.57	28	5.76
9	Pneumococcus culture broth, avirulent strain, 10-day culture	0.1	0.2	30	0.1	25	8.33	6	0.50
10	Diphtheroid culture broth, 18-day culture, pH 8.2+	0.1	0.2	30	0.1	25	8.50	8	0.68
11	B. fecalis alkaligenes culture broth, 18-day culture, pH 7.8	0.1	0.2	30	0.1	25	5.75	8	0.46
12	Staphylococcus aureus culture broth,[5] 18-day culture, pH 8.2	0.1	0.2	30	0.1	25	5.00	2	0.10
13	B. Hofmanni culture broth, 18-day culture, pH 7.8	0.1	0.2	30	0.1	25	15.00	6	0.90
14	B. coli culture broth, 18-day culture, pH 8.2	0.1	0.2	30	0.1	25	20.50	8	1.64
15	B. subtilis culture broth, 18-day culture, pH 8.2	0.1	0.2	30	0.1	25	15.71	14	2.20
16	B. diphtheriæ culture broth, Park No. 8 strain[6] x, 18-day culture, pH 8.2+	0.1	0.2	30	0.1	25	7.00	14	0.98
	Control broth, incubated 18 days, pH 7.45	0.1	0.2	30	0.1	25	16.40	64	10.50
	Control broth, not incubated, pH 7.45	0.1	0.2	30	0.1	25	14.45	74	10.70
	Control, 0.85 per cent salt solution	0.1	0.2	30	0.1	25	14.02	72	10.10

[1] Standard laboratory toxin.

[2] From an attenuated strain of Park No. 8.

[3] Toxin produced at room temperature.

[4] These lines of investigation were originally started with the pneumococcus in 1907, but when they were resumed attention was directed to the study of the better-known toxins. The pneumococcus culture broth also has marked action on the red blood cells, which is easily seen when it is added in excess; the mixture turns brown and the red cells may be laked. The leukocytes, however, persist and a few of them continue to be active phagocytes. The pneumococcus cultures used in this protocol were originally the same Type I strain. The avirulent culture was obtained by attenuating the virulent strain in culture media (see A Study of the Changes in Virulence of the Pneumococcus at Different Periods of Growth and under Different Conditions of Cultivation in Media, by A. B. Wadsworth and M. B. Kirkbride, Jour. Exper. Med., 1918, xxviii, 791–805). The avirulent culture grew much more luxuriantly than the virulent culture but often in repeated tests the action of cultures of both strains on the leucocytes varied greatly.

[5] Same strain as that used to test phagocytosis in all tests.

[6] Same strain as that used for standard toxin No. 161.

NOTE.—Salt solution controls are not included, since the results were always practically the same as those of the broth controls. The broth controls were in all cases from the same lot of broth as that in which the organisms were grown.

In the second series of experiments some facts regarding the nature of this depressing substance in toxins and culture broths were determined.

Diphtheria toxin was generally used and the technic was the same as that for the first series. The tests readily fell into four groups: (1) The effect of the time of exposure, (2) the effect of neutralizing agents, (3) the effect of destructive agents, and (4) the source of the depressing factor.

1. The Time of Exposure. Leukocytes were exposed to the action of diphtheria and tetanus toxins for varying lengths of time between the limits of two or three minutes and seven hours. Then staphylococci were added for phagocytosis. The degree of phagocytic depression, however, bore no relation to the time of the exposure of the leukocytes. The action of both of the toxins was immediate, and there was no evidence that the leukocytes later recovered in any degree by neutralizing the primary effect.

2. Neutralizing Agents—Antiserums. Diphtheria toxin-antitoxin mixtures were made with standard laboratory products as follows: (*a*) Toxin with no antitoxin, (*b*) toxin with antitoxin to $\frac{1}{100}$ neutralize, (*c*) toxin with antitoxin to $\frac{1}{10}$ neutralize, (*d*) toxin with antitoxin to just neutralize, (*e*) toxin with antitoxin to several times neutralize, and, as usual, all the necessary controls—normal dog serum, normal horse serum, undiluted antitoxin, broth and salt solution—were included. These mixtures were left at 37° C. for varying lengths of time between the limits of two or three minutes and seven hours. Leukocytes were then exposed to the action of these test mixtures for fifteen minutes before the staphylococci were added. The resulting indices showed that the antitoxin had no neutralizing effect on the depressing element in the toxin nor did the toxin in small amounts act as a stimulant to the phagocytes.

Unconcentrated diphtheria antitoxic horse serum was allowed to act upon an equal volume of its homologous toxin for one hour. Leukocytes exposed to this test mixture for fifteen minutes gave as depressed indices as the toxin control with no antiserum.

An agglutinating horse serum of high titer, produced by immunization with twenty-four-hour cultures of diphtheria organisms washed free of all toxin, was used as a possible neutralizing agent for the

phagocytic depressing substance in diphtheria toxin of the homologous organism. The attempt was entirely unsuccessful. The agglutinating serum possessed no neutralizing elements.

3. Destructive Agents—Light, Heat and Enzymes. Standard diphtheria and tetanus toxins were exposed to diffuse daylight for fourteen days and then to direct sunlight for three and one-half hours without in any way lessening the depressing effects of those toxins on the phagocytic action of the leukocytes.

The diphtheria, tetanus and botulinus toxins and tuberculosis culture broth described in the table were subjected to varying degrees of heat. The upper limit was 120° C. for one hour under fifteen pounds pressure in an autoclave. This heating in no instance produced the slightest effect upon the depressing element.

Dilute solutions of two proteolytic enzymes, trypsin and pepsin, in 0.85 per cent salt solution were allowed to act upon tetanus, diphtheria and botulinus toxins and culture broths of the tubercle bacillus and of the staphylococcus for eight hours at 37° C. Leukocytes exposed to these digested bacterial toxins and culture broths as test substances for fifteen minutes before adding the staphylococci showed a much higher degree of phagocytic activity than the undigested toxin controls, and in some cases the depressing substance was completely destroyed. The only instance in which no neutralization was apparent was that of trypsin in botulinus toxin. Possibly eight hours digestion was inadequate. Further work should be done on this point.

4. Source of the Depressing Factor—Culture Medium. Diphtheria cultures of Park No. 8 organisms were grown in several experimental synthetic culture media for toxin production. The minimal lethal dose of the resulting toxins varied from 0.005 cc. to 0.1 cc. according as the constituents of the media were varied.[1] When the action of these toxins was tested on leukocytes it was found that the composition of the medium—which is most important in the production of standard diphtheria toxin—in no way affected the production of the element which depressed the phagocytic index. All of the toxins were equally depressing.

[1] These toxins were produced in this laboratory by Mary Wheeler in an investigation of the conditions affecting the production of the bacterial toxins.

The age of the cultures was found to be important in producing the depressing substance. This, however, varied with the organism; a three-day culture broth of *Bacillus Hofmanni* depressed the phagocytic index only 21 per cent while that of staphylococcus caused a reduction of 55 per cent. The standard diphtheria toxin was cultivated seven days. The age of the toxins and culture broths used is included in the second column of the table.

The method of freeing the culture broths of organisms was immaterial—whether by centrifugalization or by Berkefeld filters.

This depressing substance has already been isolated from standard diphtheria toxin by adsorbing it to leukocytes and then lightly washing it from them with physiological salt solution. The leukocytes so treated regained their normal phagocyting ability after the washing, the toxin used ceased to depress the phagocytic activity of normal leukocytes and the salt solution washings took on the depressing property of the toxin for leukocytes in a phagocytic mixture.

Summary of Experimental Results. To sum up the experimental section: First, the action of culture broths of thirteen widely differing pathogenic and saprophytic bacterial species was tested on phagocytes *in vitro*. In every case the phagocytic power of the leukocytes was inhibited in a high degree. Second, tests, chiefly with a standard diphtheria toxin, were done to determine some facts concerning the nature of this substance depressing to phagocytic activity and its relation to the true toxins. These tests showed that its action was immediate and could not be neutralized by the ordinary antiserums tested nor destroyed by exposure to the degrees of heat or light used in the experiments. Variations in the constitution of the culture broths, which greatly affected true toxin production, caused no variation in the production of the depressing substance. The depressing action of young culture broths was found to be less marked than that of older cultures. It was also found that digestion with proteolytic enzymes either wholly or partially destroyed the depressing element. The substance could be isolated by adsorbing it to leukocytes and then washing it from them with salt solution. After removal of the substance the leukocytes regained their phagocytic activity.

STUDIES IN DIABETES INSIPIDUS, WATER BALANCE, AND WATER INTOXICATION

Study I

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AND
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Two years ago we developed a special interest in diabetes insipidus, and as a result have been carrying out investigations on patients suffering from this disease. During the course of clinical studies certain observations were made which led to experiments in animals relative to water balance and to water intoxication.

In this study we shall report (1) the results of our clinical studies in the fifteen cases of diabetes insipidus observed during the last two years in the Mayo Clinic and in the University Hospital, Minneapolis; (2) the effects on urinary secretion of the subcutaneous injections of the extract of the posterior lobe of the pituitary gland in normal dogs and in dogs which have undergone resection of the renal nerves; and (3) the production of water intoxication by the administration of water by mouth subsequent to the subcutaneous administration of pituitary extract.

Studies in Diabetes Insipidus[1]

Etiology. We have made an attempt to determine the etiology of the diabetes insipidus in our cases. One patient had brain

[1] The details of the clinical studies will appear in a separate publication by Dr. E. E. Larson.

tumor, not definitely localized but known to be supratentorial, another hypophyseal tumor and four had syphilis; in nine no causative factor could be determined. The incidence of syphilis in our series is high. Its etiologic importance has been emphasized by Fournier, Futcher and Oppenheim. Whether or not syphilis was responsible for the diabetes insipidus in our cases could not be determined, but vigorous treatment for syphilis failed to exercise any significant effect on the course of the diabetes insipidus.

The Nature of the Disturbance of Water Balance. The cardinal symptoms of diabetes insipidus are related to disturbance of water metabolism; water is ingested and excreted in amounts far in excess of normal. In Trousseau's case, for example, the most extreme on record, the fluid intake amounted to 40 liters and the urinary output to 43 liters a day. Thirst, the most annoying symptom, occasions extreme discomfort at times, while polyuria and polydypsia are the source of great inconvenience. Constipation and absence of sweating minimize the loss of fluid from the bowels and skin. The exact amount of fluid lost by way of the lungs and skin have not been determined accurately in diabetes insipidus so far as we know. Such studies are most desirable, but they necessitate the employment of special apparatus. Loss of fluid by way of the lungs plays a relatively smaller part than in the normal person. Studies of fluid balance in our cases show a striking correlation between the fluid intake and the urinary output. The literature abounds with similar results, but some writers have recorded urinary excretion somewhat in excess of the fluid ingested.

The fundamental question of whether the thirst or the polyuria is primary we find difficult to answer. In this connection we have considered factors as follows: (1) The sequence of onset of symptoms, that is, whether thirst or polyuria constitutes the initial symptom; (2) the influences of various procedures in controlling thirst and urinary output; and (3) the presence or absence of organic or functional changes in the urinary tract.

Sequence of Symptoms. By careful questioning with regard to the onset of the disease it was possible, in three of our cases, to elicit a definite history of sudden onset of thirst and to establish that in two thirst was primary, that is, it preceded the polyuria. In the

first case the circumstances surrounding the onset of the disease were such as to make the evidence almost conclusive. The patient, a farmer's wife, was left alone in her home throughout the entire evening. She became extremely nervous and was frantic with fright. She lay in bed most of this time too frightened to move. During this period she developed a terrific thirst. She suffered no discomfort from bladder distention, nor did she void. She states that on the return of the family she rushed to the spigot and drank copiously of water, taking about two quarts. She insists that she did not void at this time, for in order to do so she would have been compelled to pass through several dark vacant rooms, which in her nervous state she would not have attempted. Polyuria developed on the following morning. The second patient, an intelligent man, insists that thirst was the primary symptom, and that it developed during the night. He got up and drank copiously of water, voided but little and did not suffer from any discomfort from bladder distention. In the third case both thirst and polyuria came on suddenly, but the patient was unable to recall the order of their appearance. In the remaining twelve cases the onset of the disease was insidious and consequently no conclusions could be reached concerning the priority of the symptoms.[1]

The Influence of Various Procedures in the Control of Thirst and Urinary Output. Cannon explains normal thirst on the basis of a local sensation resulting from local dryness of the buccal mucous membrane due to decreased secretion from the salivary glands, which in turn is dependent on the diminished supply of fluid furnished these glands by the body because of its depletion in water. Cannon further states that the osmotic pressure of the blood remains unchanged despite deprivation of fluids in the tissues. Dehydration of all tissues, including salivary glands, leads to diminished secretion of water by these glands. Normally the salivary glands furnish the index to the body need for water.

Thirst and a dry mouth were complained of by all our patients.

[1] Since this paper was written Bailey and Bremer have reported their studies in experimentally induced diabetes insipidus before the Society of Endocrinology, Boston, June, 1921. They have demonstrated conclusively that thirst and polydypsia may precede polyuria.

Two volunteered the information that they experienced a peculiar sensation "as though the mouth were full of cotton." One demonstrated that "tenacious strings of saliva stretched from the roof of the mouth to the tongue when the mouth was opened wide." There is little doubt that the viscidity of the saliva is increased.

We attempted to determine the role of the local sensation in the mouth with regard to water intake and urinary output in diabetes insipidus. We believe that specific local nervous influences have been excluded as a cause of the thirst, since cocainization of the mucous membrane of the mouth and of the nasopharynx to the point of anesthesia failed to control either the polydypsia or the polyuria. In fact, in one case cocainization was pushed to the point of mild constitutional toxicity with no effect on the diabetes insipidus. In other cases pilocarpin was injected to procure salivary secretion, but without results; in four of these cases there was no evidence of decreased urinary output nor of diminution of thirst. As a result of these studies it appears that the thirst in diabetes insipidus is more than the mere expression of dryness of the oral mucous membrane.

The Presence or Absence of Organic and Functional Disturbances in the Urinary Tract. Careful studies of renal function in diabetes insipidus failed to incriminate any part of the urinary tract as an etiologic factor and revealed no deviations from normal function except in relation to water excretion and perhaps to salt excretion.

In all our cases the urinary output was markedly increased, from 3 to 14 liters a day. The specific gravity was markedly decreased, ranging, as a rule, between 1.001 and 1.004. Albumin appeared intermittently in small amounts in three cases while demonstrable glycosuria was persistently absent. The excretion of phenolsulphonephthalein and the values for blood urea, creatinin, and uric acid were uniformly normal. Cystoscopic examination in two cases showed normal bladders. Ureteral catheterization revealed the fact that the increased secretion of urine was bilateral and approximately proportional for both kidneys, and that the appearance time for phenolsulphonephthalein was normal from each side. Administration of pituitary extract with the ureteral catheters

in place, reaching to the pelvis of the kidney, resulted in control of the polyuria, and in a change in the concentration and color of the urine within eight minutes in one case and within ten minutes in another. These results conclusively exclude constriction of the ureter as a factor in the control exercised by pituitary extract on the polyuria and gross organic changes in the kidney and urinary tract. Hoppe-Seyler presents the situation aptly when he says of renal function in diabetes insipidus, "It fails only by a little pituitrin of being normal."

The Results of Treatment. The discovery by Schäfer and his colleagues of the influence of the extract of the posterior lobe of the pituitary on the urinary output has created a new and growing interest in the subject of diabetes insipidus. Unfortunately Schäfer's observations were made on anesthetized animals and led to the belief that the active substance of the posterior lobe possesses diuretic properties. Physiologists and clinicians were alike deceived with the result that diabetes insipidus was looked on by many as the result of overactivity of the posterior lobe. However, in 1913, von der Velden in Germany and Farini and Ceccaroni in Italy, working independently, administered extract of the posterior lobe to patients suffering from diabetes insipidus and obtained an effect almost specific; thirst, polydypsia and polyuria were all promptly and effectively controlled, at least for a temporary period.

Data Concerning the Fifteen Cases in the Series

The Effect of Pituitary Extract. The effect of pituitary extract was studied in all our cases. Marked temporary results were obtained in all. The effects were immediate and lasted for periods ranging from a few hours to four or five days (Charts 1, 2 and 3). In mild cases the water balance was reduced to its normal level while in more severe cases the level was reduced strikingly. Pituitary extract in gum acacia exercised as good an effect and resulted in a somewhat more prolonged control, although the local reaction was somewhat more severe. Effects of short duration sometimes follow the administration of pituitary extract by rectum.

With the decrease in the amount of urine the specific gravity rose

to 1.010 or 1.025 and the color increased proportionately. With the readjustment of urine secretion the saliva flowed more freely; the

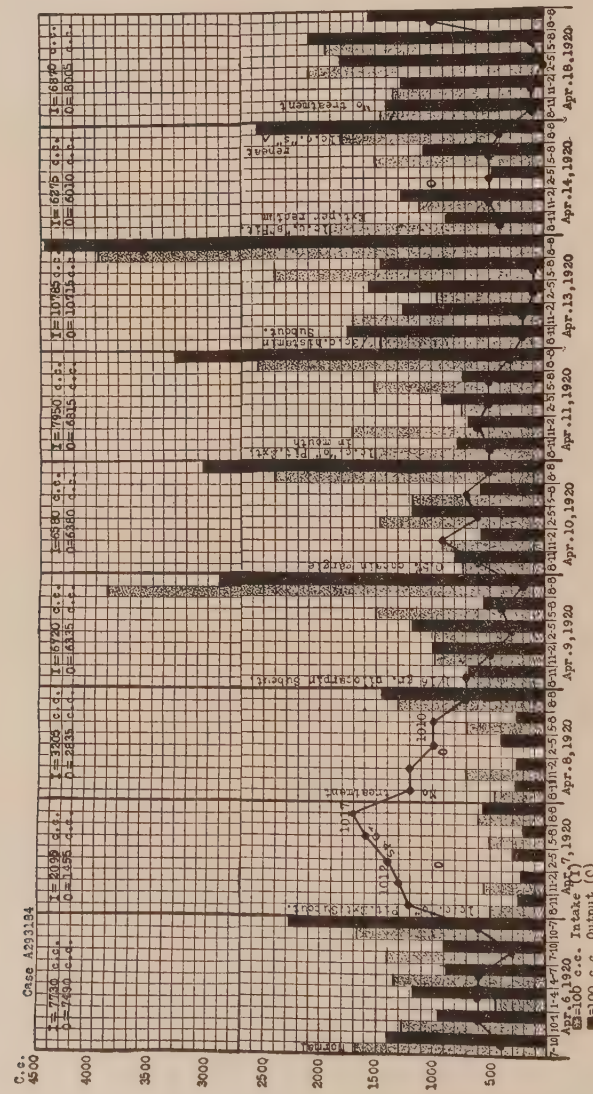

CHART 1.—Case A 293184. Fluid intake and urine volume and specific gravity in the three-hour periods while the patient was receiving varying treatment.

skin became moist and actual perspiration occurred in a large number of the cases. With the diminished desire for water the

taste for it was frequently perverted, the patients referring to water at these times as being "stale" or "flat." A sensation of satiety and a feeling of fullness resulted from the intake of small quantities of water, for example, half a glass. This is in striking contrast to

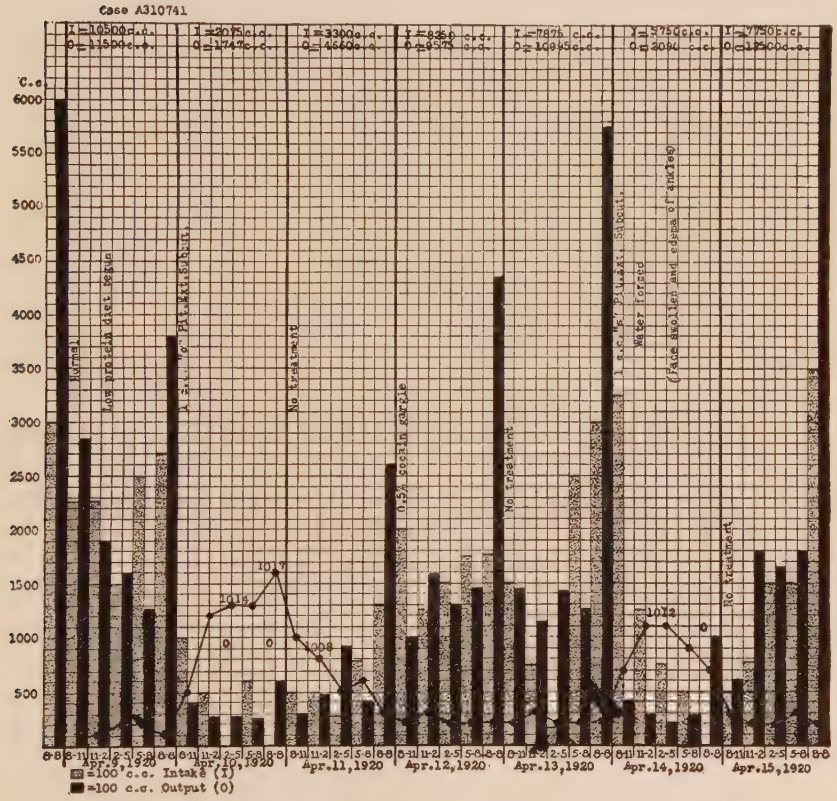

CHART 2.—Case A 310741. Fluid intake and urine volume and specific gravity in three-hour periods during marked antidiuretic effect of pituitary extract and water intoxication.

their usual experience. Prior to the administration of pituitary extract a quart of water often failed to relieve the consuming thirst.

The volume of saliva secreted after the administration of pituitary extract increased definitely. This is illustrated in Chart 4, which

contrasts the action of pituitary extract on the volume of urine and saliva of normal persons and of a patient with diabetes insipidus.

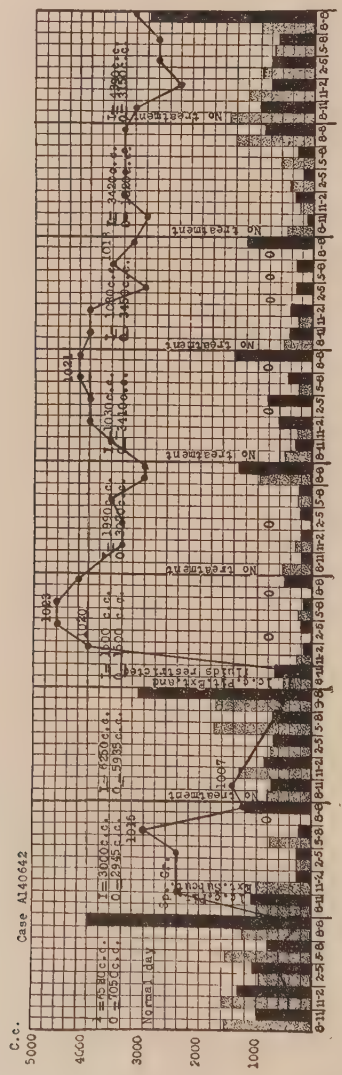

CHART 3.—Case 140642. Prolonged antidiuretic effect of pituitary extract when patient exercised voluntary restriction on his thirst.

EFFECTS OF HISTAMIN. Histamin was administered subcutaneously in three cases. Constitutionally and locally its effect dif-

fered entirely from that of pituitary extract. In no instance was either the thirst or the polyuria decreased, except on one occasion when slight control was noted for three hours. The usual type of result may be seen in Chart 1.

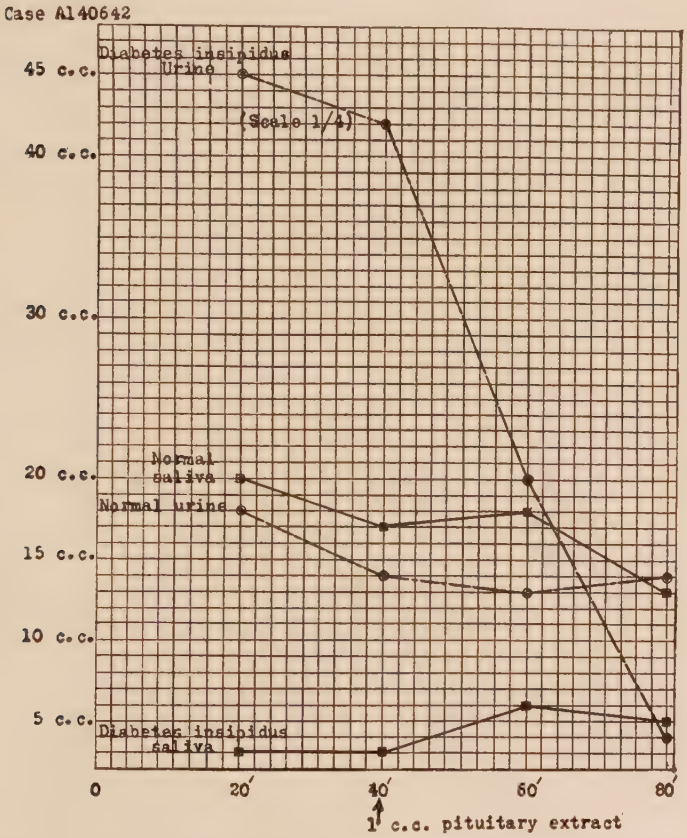

CHART 4.—Case A 140642. Effect of pituitary extract on salivary and urinary secretion.

RESULTS OF SPINAL PUNCTURE. A spinal puncture with the withdrawal of from 5 to 10 cc of cerebrospinal fluid was performed in six cases without demonstrable effect on any of the cardinal symptoms. Because of the favorable reports in the literature we believe that the amount of fluid withdrawn might be of significance.

Consequently spinal drainage was done in three other cases, but in not a single instance was a favorable effect obtained.

Results of Treatment for Syphilis. In the four cases with evidence of syphilis, treatment was carried out, but it has proved unavailing to date. Spinal puncture in three of these cases also failed to give relief.

The Effect of Restriction of Fluids. Voluntary restriction of fluids was attempted in several of our cases, but it met with minor success in two only. One patient, a woman, succeeded in decreasing the intake from 5 to 6 liters a day to 1.5 liters, through her determined effort to refrain from water. Her case was unusual, however, the diabetes in all probability being hysteric in nature. In the second case early syphilis of the central nervous system complicated the diabetes insipidus. By the aid of 1 cc of pituitary extract restriction to approximately normal amounts was obtained for a period of five days (Chart 3). Without pituitary extract attempts at restriction accomplished but little, while pituitary extract alone failed to exercise control for periods longer than two days.

Before prescribing restriction of fluids the intense distress accompanying water-fasting should be duly considered. One of our patients who had undergone a "restriction cure" at the hands of her local physician had tears streaming down her face as she told us her story. In true diabetes insipidus little or nothing can be accomplished through restriction of the fluid intake and the suffering inflicted is out of all proportion to the slight benefit obtained.

Metabolism.[1] In a consideration of metabolism in diabetes insipidus the changes induced in normal persons by copious water-drinking must be kept in mind, as has been pointed out by several authors. Rosenbloom and Price give an excellent review of the literature on the metabolism in this disease. Gibson and Martin have recently published results of metabolic studies before and after administration of pituitary extract.

Metabolism studies have been carried out in two cases of diabetes insipidus (Cases 140642 and 330908), the study in one being repeated during the patient's second visit to the Clinic. Studies of blood

[1] The details of these metabolic studies will appear in a separate publication by Dr. J. F. Weir.

chemistry were made in two other cases (Cases 317545 and 323015) before and after the administration of pituitary extract. The results in one case (Case 330908) are shown in Tables 1, 2 and 3.

TABLE 1.—URINE ON METABOLIC EXPERIMENT (A 330908)

Day.	Volume, cc.	Average specific gravity.	N/10 acid, cc.	Chlorin, gm.	Total nitrogen, gm.	Urea nitrogen, gm.	Ammonia nitrogen, gm.	Creatinin nitrogen, gm.	Uric acid nitrogen, gm.	Urea nitrogen, per cent total.	Ammonia nitrogen, per cent total.	Creatinin nitrogen, per cent total.	Uric acid, per cent total.
						BEFORE TREATMENT							
1	4525	1.007	398	3.450	8.260	6.560	0.316	0.316	0.092	79.4	3.80	3.8	1.1
2	4980	1.009	343	4.300	8.020	..	0.266	0.511	0.048	..	3.30	6.4	0.6
						DURING TREATMENT							
3	915	1.025	268	2.590	8.134	5.497	0.466[1]	0.493	0.071	67.5	5.70	6.1	0.9
4	1200	1.024	356	3.400	8.200	7.092	0.209	0.537	0.120	86.5	2.50	6.5	1.5
						AFTER TREATMENT							
5	4445	1.018	318	7.190	9.270	7.998	0.309	0.501	0.116	86.4	3.30	5.4	1.2
6	5960	1.006	419	5.721	8.020	6.385	0.308	0.475	0.117	79.6	3.81	5.9	1.5

[1] Urine voided, alkaline at one period on this day.

TABLE 2.—BALANCE OF WATER, NITROGEN, AND CHLORIN (A 330908)

Day.	Intake.			Urine output.			Balance.			Weight, pounds.
	Water, cc.	Nitrogen, gm.	Chlorin, gm.	Water, cc.	Nitrogen, gm.	Chlorin, gm.	Water, cc.	Nitrogen, gm.	Chlorin, gm.	
1	5189	8.42	5.94	4525	8.26	3.45	+ 664	+0.16	+2.49	162.5
2	5740	8.22	6.38	4980	7.96	4.30	+ 760	+0.26	+2.08	162.5
3	3193	7.78	5.44	915	8.14	2.59	+2278	−0.36	+2.85	163.0
4	2380	7.92	6.27	1200	8.20	3.40	+1180	−0.28	+2.87	164.7
5	4732	8.61	5.62	4445	9.27	7.19	+ 287	−0.66	+1.57	164.2
6	6382	7.57	5.80	5960	8.02	5.72	+4422	−0.45	+0.08	162.0

The water intake showed a marked fall in the three hourly and daily quantities after the administration of pituitary extract. The variations in the volume and specific gravity of the urine were marked and obvious. The positive water balance in the control period was small, harmonizing with the clinical observation of dry skin and mouth. There is marked retention of water after the first dose of pituitary extract, corresponding with the decreased urinary

output. The patients generally report an increased excretion of urine in the period after treatment. In this case, however, the water intake kept pace with the increased excretion, but this fact, together with the increase in weight during treatment and the return to normal in the after period, indicates retention of water in the blood or tissues. The excretion of nitrogen varied but slightly throughout the experiment and the nitrogen balance showed no appreciable change. The chlorin excretion was decreased under treatment and chlorin retention was evident.

TABLE 3.—BLOOD ON METABOLIC EXPERIMENT (A 330908)

Day.	Hematocrit, per cent plasma.	Degrees.	Plasma, mg. per 100 cc.		Whole blood, mg. per 100 cc.					Remarks.
			Total nitrogen.	Sodium chlorid.	Total non-protein nitrogen.	Urea nitrogen.	Uric acid.	Creatinin.	Sugar.	
					BEFORE TREATMENT					
1. . .	67	0.515	1171	648	29.8	14.9	2.68	1.90	128	Normal.
2. . .	68	0.530	1142	653	26.5	14.9	..	..	108	Normal.
					DURING TREATMENT					
3. . .	69	0.542	1082	640	26.6	16.2	..	..	144	Pituitary extract.
4. . .	72	0.531	1060	623	24.8	14.8	..	..	108	Pituitary extract.
					AFTER TREATMENT					
5. . .	71	0.522	1099	633	28.1	14.8	..	..	108	After period.
6. . .	69	0.546	1129	648	27.0	13.3	..	..	112	After period.

In the second case (Case 140642) similar changes in the urine were noted and more marked change in the water balance; there was a large negative balance as the effects of the drug wore off. There was a definite decrease in the excretion of nitrogen and an increased positive balance in the period of treatment. Chlorin also showed rather marked retention.

In the analysis of the blood in diabetes insipidus before and after administration of pituitary extract certain definite changes appeared. Typical results in one case (Case 317545) are contrasted with those of a normal person (Table 4). The relative plasma volume is increased and the molecular concentration, the total nitrogen and

the sodium chlorid of the blood are somewhat decreased, findings which might indicate a dilution of the blood. However, in the findings in the experiments on metabolism there is evidence of a slight retention of nitrogen and chlorin which does not appear in studies on the blood. Evidently water retention is the most prominent feature following administration of pituitary extract, and in all probability the effects on nitrogen and chlorin are secondary to water retention.

TABLE 4.—EFFECT OF PITUITARY EXTRACT IN NORMAL PERSON AND IN PATIENT WITH DIABETES INSIPIDUS CONTRASTED IN BLOOD PICTURE

Day.	Hematocrit, per cent plasma.	Degree.	Normal.							Remarks.
			Plasma, mg. per 100 cc.	Whole blood, mg. per 100 cc.						
			Total nitrogen.	Sodium chlorid.	Total non-protein nitrogen.	Urea nitrogen.	Uric acid.	Creatinin.	Sugar.	
1. . .	65	0.523	880	600	42	..	..	..	..	Control.
2. . .	58	0.534	1008	605	43	16.0	..	..	146	Control.
3. . .	60	0.533	977	610	40	15.0	..	..	152	Pituitary extract.
4. . .	61	0.534	942	618	37	13.0	..	..	149	Pituitary extract.
			Diabetes insipidus (A31 7543)							
1. . .	61	0.522	1058	629	..	5.0	2.4	1.3	..	Control.
2. . .	55	0.549	1163	640	..	5.8	2.4	1.3	..	Control.
3. . .	67	0.511	940	607	..	6.6	2.4	1.3	..	Pituitary extract.
4. . .	67	0.513	970	597	..	6.1	2.3	1.3	..	Pituitary extract.

In all our experiments on metabolism and in all our cases of diabetes insipidus in which pituitary extract has been administered there has been no diarrhea,[1] and an analysis of the feces in one case showed a normal quantity of water. There is no evidence that pituitary extract prevents the absorption of water from the gastro-intestinal tract of man, as has been claimed for it for rabbits by Rees. Only small quantities of water have been found in the intestines and feces of our animals at necropsy.

[1] Diarrhea was present in animals suffering from water intoxication, but the amount lost by way of the bowels was relatively very small.

Blood Volume. Blood volume determinations by the vital red method were carried out in four of our patients, and in three of them before and after the administration of pituitary extract, that is, in the periods of high and low urinary excretion. The results obtained in the two earlier cases led us to believe that the blood volume, particularly the plasma volume, was increased in diabetes insipidus following the administration of pituitary extract, that the pituitary extract had specifically raised the renal threshold for water and that the kidney failed to excrete water even in the presence of hydremic plethora in the blood. However, in one case no increase was found in either total blood volume or plasma volume, despite the completeness of control exercised by the pituitary extract on the polyuria. Prior to the administration of the drug the plasma and total blood volume were 3460 cc and 4805 cc respectively, and after the drug 3495 cc and 4855 cc respectively. From these results it is evident that pituitary extract can control the polyuria of diabetes insipidus and prevent polyuria following excessive ingestion of water without the development of any significant increase in blood or plasma volume. These findings are in keeping with the experimental results which we obtained in animals.

The Effects of Pituitary Extract on the Water Balance

Behavior and Role of Water in Metabolism. In order better to understand the effect of the pituitary extract on the water balance it is desirable to know more concerning the nature of water exchange in the body, the mechanism involved in its regulation and the part played by the pituitary gland under normal and abnormal conditions. Water plays a fundamental part in metabolism and is essential to the life and function of every living cell. It is transported to and from the cells by the blood, provision for its exchange being made locally by the presence of lymph spaces and lymph channels. It constitutes 80 per cent of the blood. Oxygen and food are also transported to the cells by the blood. Nature has arranged to supply the blood with water, oxygen and food, according to the constancy of the need of the cells for each and in accordance with

the complexity or simplicity of the mechanism involved in their metabolism. Water holds a position intermediate between oxygen and food, the continuation of its supply being less vital than that of oxygen and more than that of food. In health blood varies in volume and consistency within narrow limits only, possibly in order that the supply of these essentials to the cells may be kept almost constant. Fluctuations in physiologic needs, depending on variations in function, are met largely by changes in the rate and volume of the blood flow to various organs.[1]

TABLE 5.—SUPPLY AND OUTPUT OF WATER

		Amount.			Author.	Remarks.
		Average.	Maximum.	Minimum.		
Supply.	Water from drink and food	....	3500	2300	Forster	Data collected in Munich; fluid includes 1 to 2 liters of beer.
		2290	2440	880	Atwater and Benedict	Over period of forty-nine days with subject in repose.
		3700	4550	2225		Over period of sixty-six days with subject doing moderate work.
	Water from oxidation of food	....	480	240		On mixed diet of approximately 4000 to 2000 calories respectively.
Output.	Urine	1500 2000	3000	800	Emerson	So many factors affect the urinary output that only a general average is given here.[1]
	Skin	550	....	....	von Noorden	These are average figures for patient indoors on a medium diet of 1800 to 2000 calories; total fluid lost by evaporation may reach 1600 cc on diet of 3500 calories and 3250 cc on diet of 5000 calories with patient doing moderate or heavy work; as much as 7.5 liters may be lost during excessive work.
	Lungs	400	....	....		
	Feces[2]	60 to 300	300	60		On vegetarian diet with large stools the water content may reach 300 cc; in diarrhea greater losses may occur.

[1] In 5 cases followed by us during summer months the output varied from 750 to 1200 cc. The figures for maximum and minimum simply indicate ordinary physiologic limits.
[2] Obviously this refers to normal loss in the stools in diarrhea.

The source of the water of the body is found in the fluids taken by mouth in the form of food and drink and in the water which results from oxygenation of food within the organism. Water is excreted

[1] The intermediary water metabolism and the effects exercised by water ingestion are purposely omitted from discussion.

by the skin, lungs, kidneys and bowels. The intake and output vary tremendously with the life, habitat, and habits of the individual. The relative amount excreted through the various channels also varies extremely, reciprocal relationships playing a striking part at times, particularly with regard to sweat and urine. Although the variations are extreme, some idea concerning the normal relations in water balance, that is, in the intake of fluids and in the output of water by the various channels under differing conditions of diet and activity, are presented in Table 5.

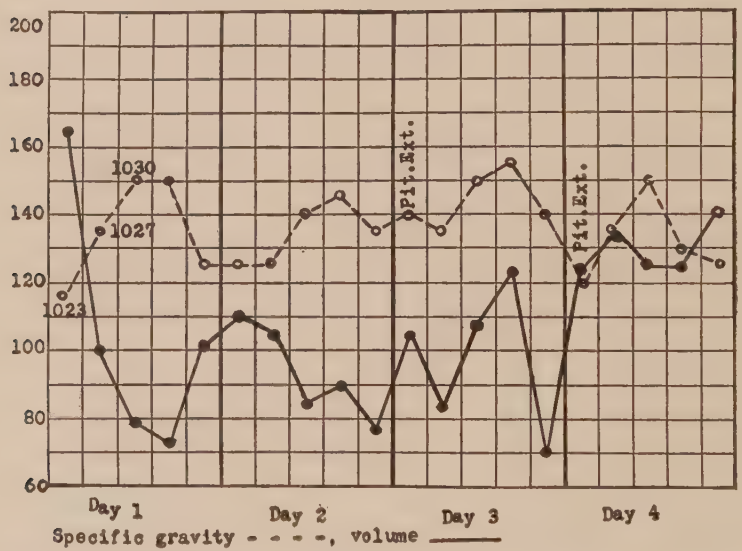

CHART 5.—Average three-hour volume and specific gravity of the urine of five normal men before and after pituitary extract.

EFFECT OF EXTRACT OF THE POSTERIOR LOBE OF THE PITUITARY GLAND ON THE URINARY EXCRETION IN NORMAL MEN AND ANIMALS. In order to ascertain the effects of pituitary extract on the urinary excretion of normal persons five men were placed under careful observation. Urine was collected in three-hour periods throughout the day and as a single specimen at night. Diet and climatic conditions remained fairly constant during the period of observation, August, 1920. A two-day period of control preceded the administration of the drug, which was given at 8 A.M. on each day of the

experiment. A composite curve in the urinary output and the specific gravity is shown in Chart 5. From these studies it is evident that the subcutaneous injection of 1 cc of pituitary extract produced no appreciable effect on the urinary excretion either on the daily or three-hour amounts or on the specific gravity.

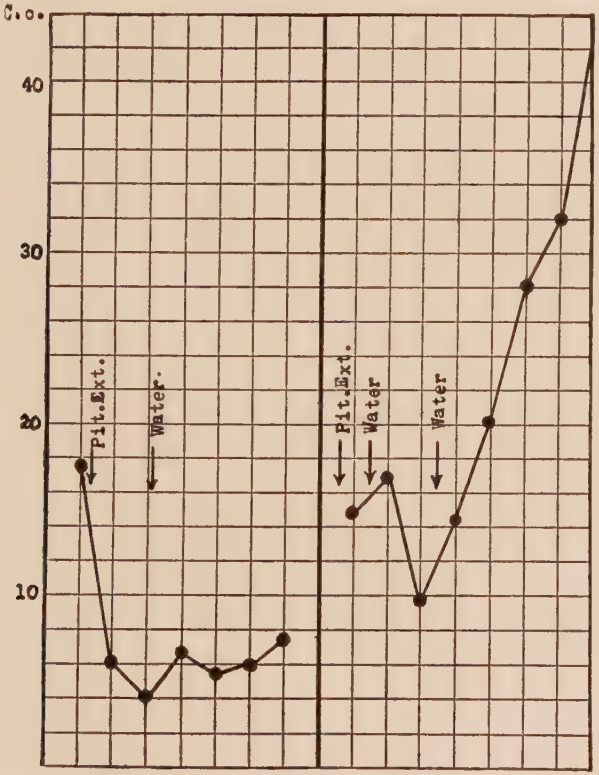

CHART 6.—The antidiuretic effect of 1 cc pituitary extract in dogs can be overcome by repeated administration of water.

Other experiments were carried out on dogs; the water was given with pituitary extract and in some instances repeated at varying periods after the administration of the drug, in order to determine if diuresis could be induced. Characteristic curves are reproduced in Chart 6. It is evident that diuresis can be readily induced by forcing the water intake after a single dose of 1 cc of pituitary extract.

In Chart 7*a* is a composite curve of the results of a series of experiments in dogs, in which the urine was collected in half-hour periods during the course of three hours. Each animal was studied

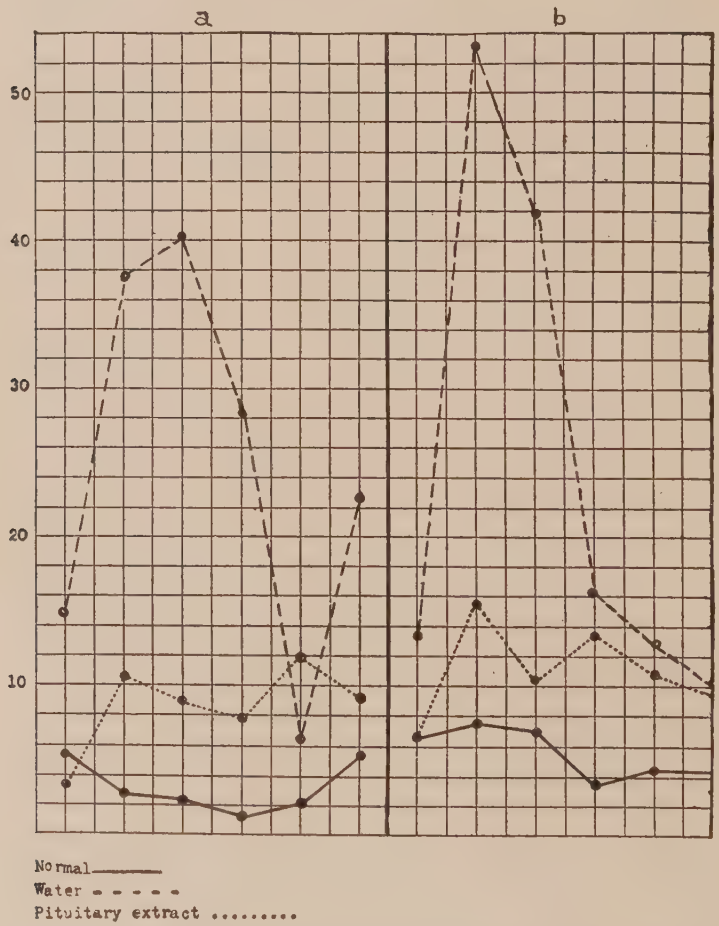

CHART 7.—Average half-hour rate of secretion of urine in dogs under normal conditions, after water and after water and pituitary extract (*a*) before section of the renal nerves (six dogs), and (*b*) after section of the renal nerves (four dogs).

under three conditions: (1) Normal conditions, (2) after administration of water by mouth in amounts corresponding to 50 cc per kilogram of body weight, and (3) after the simultaneous administra-

tion of pituitary extract subcutaneously and water by mouth in the amounts already indicated. These experiments demonstrate that the subcutaneous administration of pituitary extract definitely checks the production of water diuresis, although if the water is administered, urinary excretion is on a somewhat higher level than normal.

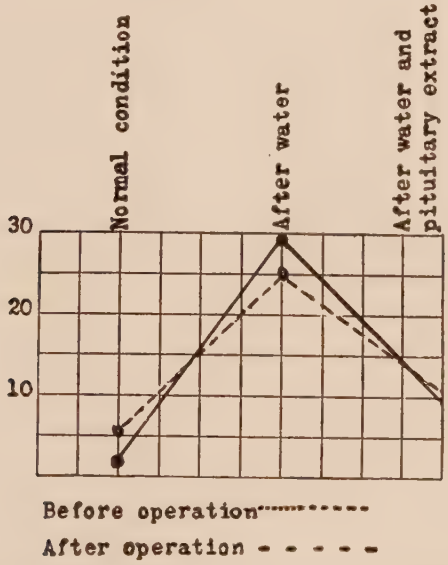

CHART 8.—Curves showing average of secretion of urine in a series of dogs under normal conditions, after water and after water and pituitary extract, before and after section of renal nerves.

The next step was an attempt to localize the site and mode of action of the pituitary extract, that is, to ascertain the relation of pituitary extract to the nervous control of renal function. The dogs of the first series were operated on and the splanchnic nerves to both kidneys sectioned.[1] A second series of experiments identical with those described was carried out (Chart 7*b*). A comparison of the total results under these two conditions is shown in Charts 7 and 8. From these two series of experiments it is possible to deduce that (1) kidneys with nerves sectioned respond normally to water with a

[1] We wish to thank Dr. F. C. Mann and his associates for their kindness in performing the operative work on the animals used in these experiments.

diuretic curve;[1] (2) this curve is largely abolished by pituitary extract, as in normal kidneys (the diuretic response after pituitary extract administration is on a slightly higher plane than under normal conditions) and (3) practically no change in the curve of the average half-hour output under the three conditions of the experiment result from section of the renal nerves. It is evident, therefore, that the influence of pituitary extract in the prevention of diuresis is independent of the nerve supply of the kidney.

The Mechanism Involved in the Control of Diuresis by Pituitary Extract. It is obvious from our clinical studies in diabetes insipidus and our experimental observations on the effect of pituitary extract in the prevention of polyuria and our investigations of water intoxication that pituitary extract prevents polyuria. The mechanism of its action is difficult to explain.

Earlier in our work we were inclined to believe that pituitary extract raised the renal threshold for water and that a hydremic plethora resulted. However, in one case of diabetes insipidus and in several animal experiments blood volume determinations failed to indicate any increase of total blood or plasma volume. As a result of our failure to demonstrate hydremic plethora and increased renal threshold for water we are considering other possibilities.

In view of the results of our studies of blood chemistry and blood volume we are at present attempting to determine if it is possible that pituitary extract disturbs the relative ease with which the water of the blood leaves the blood stream to pass through the kidney to form urine or to the tissues as body fluid. At present we are forced to admit our inability to explain the antidiuretic effect of pituitary extract. We are investigating the possibility of an abnormal escape of fluid from the circulating blood into the tissues.

Intoxication Following the Administration of Water by Mouth Subsequent to Giving Extract of the Posterior Lobe of the Pituitary Subcutaneously

One of our patients at our solicitation continued after the administration of pituitary extract to take water in amounts to which he

[1] Our results with the denervated kidney correspond to those already described by Marshall and Kolls.

had become accustomed,[1] and during a period of eight hours he ingested 5.25 liters and excreted 800 cc of urine. In the course of three or four hours he became very ill, was nauseated and developed severe headache, and was forced to go to bed. Physical examination revealed only puffiness of the lower eyelids and slight edema of the ankles. Repetition of this experiment on the same and on another patient gave similar results, that is, nausea, vomiting and headache, which forced the patient to go to bed. In one instance definite ataxia appeared.

On account of the severity of the reaction no further experiments were tried on patients but similar studies were carried out on dogs. Water in large quantities was given by mouth subsequent to the administration of pituitary extract. This resulted in marked tremor, salivation and at times vomiting. The symptoms were independent of the temperature of the water and were elicited with warm water as rapidly as with cold. On forcing the experiment convulsions and coma developed, and in one instance death supervened. As a result of these observations we undertook an investigation to determine the responsible factors and studied separately the toxicity of water and of pituitary extract, and later the toxicity of water following the administration of pituitary extract.

Toxicity of Water. In the classic experiments of Cohnheim and Lichtheim, in which they demonstrated the absence of anasarca, especially of edema of subcutaneous tissues, when animals were given physiologic salt solution intravenously in large amounts (up to 90 per cent of body weight), cramps were reported in a few instances. In an elaboration of this work by Magnus and Schäfer no mention is made of these toxic manifestations.

Miller and Williams recently have published their results following the administration of large quantities of water to patients with chronic nephritis. They observed headache, dizziness, restlessness, chills, fullness of the abdomen, vomiting, dyspnea and cramps in the legs, which were associated with marked increase in weight and definite elevation of blood-pressure. In our experiments large amounts of water were given to dogs without the development of

[1] The edema and toxicity resulting in this case were reported by one of us (Rowntree) before the Society of Clinical Investigations, Atlantic City, 1920.

untoward signs or symptoms. One animal retained 2500 cc during the course of seven hours and passed 1500 cc of urine without manifesting symptoms of intoxication. In another the administration of 3500 cc resulted in salivation, vomiting, asthenia and ataxia.

One of our colleagues, Dr. Amberg, has kindly furnished us with the following note, which is of interest in this connection. For the purpose of studying the influence of water ingestion on the elasticity of the skin as determined by the elastometer of Schade, two normal adults drank from 2 to 3 liters of water (not cold) within about fifteen to twenty minutes. For some time following the ingestion of the water no elastometric determinations could be made on account of muscular twitchings. In the light of the experiments here reported it appears not impossible that this was the result of a slight water intoxication. There were no particularly uncomfortable subjective sensations.

Toxicity of Pituitary Extract. Schäfer and Vincent state that subcutaneous injections of pituitary extract to small mammals result in paralytic symptoms similar to those observed after suprarenal extract. Cushny states that large quantities can be injected without producing symptoms other than somnolence and muscular weakness. According to Sollmann, ataxia and motor symptoms, changes in carbohydrate metabolism and emaciation develop after the excessive and long-continued use of pituitary extract.

In order to determine the etiologic relationship of the extract of the posterior lobe of the pituitary to the symptoms observed in our studies some experiments were carried out; illustrative results appear in the following protocols:

Protocol 1

Dog 1, weight 9.8 kg. Pituitrin (Park, Davis & Co.) subcutaneously. February 11, 1921. Pulse 100, regular.

10.00 A.M.	1 cc pituitary extract.
10.30 A.M.	1 cc pituitary extract.
11.00 A.M.	1 cc pituitary extract, diarrhea.
12.00 M.	1 cc pituitary extract, diarrhea.
1.00 P.M.	1 cc pituitary extract.
1.30 P.M.	1 cc pituitary extract.
1.45 P.M.	Pulse 48, sinus arrhythmia, extrasystoles; bright and interested in other animals; no salivation; respiration, 30.

2.25 P.M. 1 cc pituitary extract.
3.15 P.M. 1 cc pituitary extract.
Pulse 60, respiration 40; appeared sick; lost "pep;" no interest in other animals; sleepy; no diarrhea or bladder frequency; no ataxia.
5.00 P.M. 1 cc pituitary extract.
5.30 P.M. 1 cc pituitary extract.
5.45 P.M. Animal more sleepy; no ataxia.
February 12:
10.00 A.M. Pulse 140, regular; animal active; appears normal.
Intake from 6.00 P.M. to 10.00 A.M. 65 cc; output 300 cc.

PROTOCOL 2

Dog. 2, weight 5.4 kg. Pituitary extract (Lilly Company) intravenously.
February 16, 1921. Animal active; pulse 80, regular.
2.25 P.M. 3 cc pituitary extract; two minutes later inactive.
3.00 P.M. Sick; at times seems unable to use hind limbs; lies flat on side.
3.05 P.M. Heart-rate 44; sinus arrhythmia; restless.
3.07 P.M. Defecation.
3.13 P.M. 3 cc pituitary extract; lacks "pep"; diarrhea; pulse 60; sinus arrhythmia.
3.29 P.M. 3 cc pituitary extract; no resistance; slight tremor; appears weak.
3.31 P.M. Vomited 15 cc yellow fluid, frothy.
3.40 P.M. Pulse, 62; reflexes active;vomiting again.
3.45 P.M. 3 cc pituitary extract; several attacks of vomiting and bowel movements.
4.00 P.M. 3 cc pituitary extract; feces and vomitus bile stained; mucus present.
4.15 P.M. 3 cc pituitary extract; inactive; does not resist; pulse, 80.
4.40 P.M. Greater activity.
February 17:
10.00 A.M. Normal activity.

TOXICITY OF WATER AFTER THE ADMINISTRATION OF PITUITARY EXTRACT.[1] Subsequently animals were given pituitary extract subcutaneously and large amounts of water through a stomach tube. Blood volume determinations and studies of the blood

[1] Water introduced intravenously as physiologic salt solution of sodium chlorid results in an entirely different set of phenomena, the dogs dying with edema of the lungs.

chemistry were made before the administration of pituitary extract and again after the onset of the symptoms. The summary of the results is given in Table 6.

TABLE 6.—BLOOD VOLUME IN WATER INTOXICATION

Dog.	Weight, kg.	Pituitary extract, cc.	Intake, cc.	Output, cc.	Hematocrit, per cent plasma.	Total plasma volume, cc.	Total blood volume, cc.	Plasma per kg.	Blood per kg.	Total nitrogen, gm. per 100 cc.	Sodium chlorid, mg. per 100 cc.	Symptoms.
E 116	10.4	7 (4+3)	6400 (4000 + 2400)	2800 (1500 + 1300)	49	613	1251	59	120	0.763	480	Drowsiness, salivation, restlessness; frequent attempts at urination, vomiting, ataxia, convulsions, coma.
					51	601	1178	58	113	0.743	430	
E 291	7.4	3	1500	500	46	370	794	50	107	..	..	Drowsiness, salivation, tremor, ataxia, vomiting, convulsions, coma, involuntary evacuations of bowels and bladder.
					46	389	846	53	114			
E 290	9.0	5	5000	2000	53	618	1157	69	128	0.490	460	Preoperative: Marked frequent attempts at urination; diarrhea, drowsiness, restlessness, ataxia, tremor, twitchings, salivation, convulsions, coma.
					57	526	923	58	102	0.595	390	
E 290	8.5	3	3150	2325	55	570	1036	67	122	..	..	Postoperative; same symptoms.
					57	650	1140	76	134			

Early Manifestations of Toxicity. The first symptoms noted may possibly be due to pituitary extract alone, namely, asthenia, restlessness, frequent attempts at urination, diarrhea and vomiting. Other symptoms, tremor and salivation, next appear, possibly due to water alone. These develop usually after approximately 300 cc of water have been given per kilogram of body weight as a minimum. This stage continues for a variable period when another set of symptoms develops. The animal becomes very drowsy and later shows muscular twitchings and ataxia on standing or walking. This may be designated as the preconvulsive stage. In spite of mental dullness the animal evidences hypersensitiveness to external stimuli, such as result from picking up the stomach tube or in passing it. Such procedures may precipitate the convulsive stage.

Later Manifestations of Toxicity; Convulsions and Coma. Convulsions were characteristically epileptiform in type; first a tonic stage, followed immediately by a clonic stage. During the tonic phase the head retracted, the jaws set, respiration ceased and the animal became cyanosed. Then sudden and violent clonic spasms developed, accompanied by frothing at the mouth and involuntary bowel and bladder evacuations. During the convulsive stage the pupils were markedly dilated, contraction occurring immediately after the cessation of the clonic spasms. During or following the convulsive stage the dog usually showed running, swimming or snapping movements and at times barked and growled, as though participating in a fight. Coma or somnolence persisted for a longer or shorter period, but the animal usually recovered fully during the course of the next few hours. Passage of the stomach tube and the administration of more water brought on further convulsions, which, if continued, ended in death. Vomiting and salivation occurred usually after the administration of from 200 to 300 cc of water per kilogram of body weight. Occasionally an animal failed to manifest salivation. Some animals made frequent attempts to urinate while others apparently exhibited no evidence of bladder irritability.

Several animals in which double renal nerve section had been performed were given pituitary extract and water. The same toxic symptoms developed but no increase or decrease in the facility with which they could be produced was noted. This further indicates that the antidiuretic action of pituitary extract is independent of the nerve supply of the kidney. A protocol of one of these experiments is presented below:

Protocol 3

Dog E 249, weight 11.4 kg.[1] Water by mouth subsequent to pituitary extract subcutaneously.

February 7, 1921:

11.50 A.M. 1 cc pituitary extract and 500 cc water.
1.45 P.M. 1 cc pituitary extract and 500 cc water
2.45 P.M. 1 cc pituitary extract and 500 cc water; some frequency of micturition.

[1] Other experiments are in progress which involve more careful consideration of the weight of the animal. To date increase in weight greater than 1 kg. has not been encountered.

3.15 P.M. Has vomited 1000 cc.
3.25 P.M. Animal found in cage, unconscious, frothing at mouth, head retracted; jerking of jaw and leg muscles. On being taken from cage was unable to walk, lay on side and went through running movements; jaws still snapping. Prostration marked, movements easily restricted; semiconscious.
3.40 P.M. Involuntary bowel evacuation, liquid. Running movements stopped; attempts to walk unsuccessful.
3.45 P.M. Conscious, but lies quiet; unable to attract animal's attention; few attempts at walking movements; respiration 70; muscles rather flaccid.
3.57 P.M. Pulse 100; pupils small; twitching muscles; feet lie as if asleep; eyes open; listless; few attempts to get up; respiration 32.
3.59 P.M. Gets up, walks in circular course, lies down.
4.02 P.M. Diarrhea, hardly able to stand, does not respond to call, is not frightened.
4.06 P.M. Various attempts to rise.
4.08 P.M. Walks, seems blind, bumps into objects.
4.15 P.M. Walks again, bumps into objects; weight 11.6 kg.
4.18 P.M. Walks until exhausted; defecation.
4.25 P.M. Improving; vision returning; attention can be attracted slightly.
4.27 P.M. Pulse 108, deep respiration; frequent urinations, little or no urine.
4.45 P.M. 500 cc water; very restless, running.
4.48 P.M. Defecation.
4.50 P.M. Running less rapidly, slightly ataxic, salivated.
4.52 P.M. Running less rapidly, walks some, more ataxia, lists to left
4.55 P.M. Complete ataxia, unable to stand.
4.56 P.M. Convulsion, head retracted; tonic and clonic phases, running movements; vomited; salivated.
5.03 P.M. Convulsion.
5.04 P.M. Convulsion.
5.12 P.M. Convulsive movements.
5.20 P.M. Has been in continuous convulsions, unconscious, continuous snapping movements of jaws, pupils widely dilated.
5.30 P.M. Same condition, more convulsions, pupils contracted after convulsion, dilated during convulsion.
5.45 P.M. Died in convulsion.

Necropsy revealed early onset of rigor mortis, normal chest and lungs and slight congestion of the liver and kidneys. The stomach

contained about 65 cc of fluid and the intestine about 25 cc. No free fluid was found in the pleural or peritoneal cavities, and there was no evidence of edema of the subcutaneous tissues of the neck or of the extremities. The brain on removal appeared to be normal in every respect.

Thus we have definite evidence of toxic symptoms produced by water, when the water-secreting function of the kidney is diminished through the subcutaneous administration of 3 cc of pituitary extract.

While the experiments were in progress an attempt was made to determine the mechanism of production of the convulsions. Kymographic records were procured in some of these experiments. A typical tracing is presented (Fig. 1). Hydremic plethora as a cause was first considered, but no increase in the total blood or plasma volume by the vital red method nor any constant increase in relative plasma volume by the hematocrit could be demonstrated (Table 6).

The total nitrogen and the sodium chlorid content of the plasma showed usually a slight decrease after the onset of symptoms. The urine volume during the following twelve hours was always increased, usually to about 500 cc, indicating a rather rapid excretion of the retained fluids. Clinical evidence of edema was absent in all the animals studied.

The Cause and Significance of Convulsions and Coma. The immediate cause and the mechanism of production of these phenomena have not been determined. But it has been demonstrated that neither pituitary extract nor water alone[1] in similar or even larger amounts is capable of producing them. Edema of the brain suggests itself as a logical explanation, but we have not succeeded in proving this to our own satisfaction. Table 6 presents the plasma and blood volume values as determined before and after water intoxication. From this it is apparent that increased blood volume has been excluded in at least some of our experiments. A marked increase in blood-pressure is encountered in water intoxication. The rise is tardy in onset and gradual in development and may

[1] Since this paper was submitted for publication, further experiments have been carried out and convulsions have been produced with water alone. The results of these further studies will be published later.

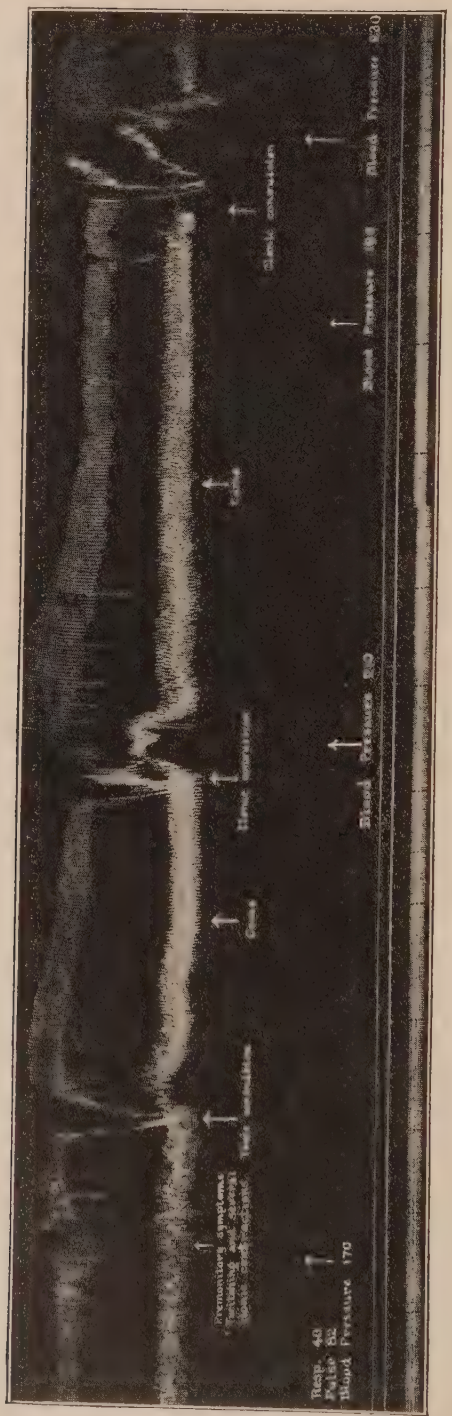

FIG. 1.—Water intoxication. Respiration, pulse and blood-pressure during convulsions in a dog.

reach a comparatively high level (Table 7, Fig. 1). However, the changes in blood-pressure do not appear to be striking enough to account for the toxic manifestations.

TABLE 7.—BLOOD-PRESSURE CHANGES IN WATER INTOXICATION

Time.	Water, cc.	Weight, kg.	Pituitrin, cc.	Blood-pressure.	Pulse.	Respiration.
11.00 A.M.	0	13.6		140	84	
11.20 A.M.	0	13.6	1 "O" subcutaneously	140		
11.30 A.M.	700	14.3		140		
11.45 A.M.	450	14.6		160		
11.50 A.M.	0	14.6	1 "O" subcutaneously	150	72	2
12.00 M.	1000	15.6		140		
1.00 P.M.	0	15.6	1 subcutaneously	140		
1.15 P.M.	1000	16.4		140		
2.05 P.M.	500	16.9	1 subcutaneously	150	70	24
3.05 P.M.	0	16.8	1 subcutaneously	170		
3.10 P.M.	500	17.2		180		
3.30 P.M.	0		1 intravenously	190		
4.00 P.M.	1000	18.1		206	72	26
4.30 P.M.	0	18.1				
4.40 P.M.	0	18.1	1 intravenously	200	82	20
4.50 P.M.	0	18.1		140	22	

Nausea, vomiting, muscle-twitching, spasms, asthenia, convulsions and coma developing in the presence of an obvious derangement of water excretion strongly suggest uremia, especially when death supervenes. But in the absence of increased values for blood urea, and in view of the rapidity of their appearance and disappearance, it does not seem at all probable. However, it is of more than passing interest to learn that the entire series of symptoms so frequently found in uremia can result from a common cause. From the standpoint of sudden onset and recovery eclampsia is also suggested.

Convulsions of sudden onset characterized by a tonic and a clonic phase, associated with involuntary evacuations and frothing at the mouth, followed by coma and ending in complete recovery, suggest epilepsy. On the other hand, nausea, vomiting, asthenia and ataxia characterizing the earlier period of development of these phenomena have no resemblance to epilepsy. The possibility of reflex factors

from the gastro-intestinal tract at least must be considered, since strangury and retching constitute marked features at times. A condition resembling status epilepticus and ending in death has also been observd.

In the absence of definite information relative to the seat, origin and mechanism of production of these phenomena speculation is futile; it would be wiser perhaps to leave all such questions to the future.

BIBLIOGRAPHY

1. Cannon, W. B.: The Physiological Basis of Thirst, Proc. Roy. Soc., London, 1917–1918, s. B., xc, 283–301.

2. Cohnheim, J., and Lichtheim, L.: Ueber Hydrämie und hydrämisches Oedem, Arch. f. path. Anat. u. Physiol., 1877, lxix, 106–143.

3. Cushny, A. R.: A Text-book of Pharmacology and Therapeutics, Philadelphia, Lea & Febiger, 1910, p. 340.

4. Farini, A., and Ceccaroni, B.: Influenza degli estratti ipofisari sull'eliminazione dell'acido ippurico, Gazz. d. osp., 1913, xxxiv, 879–882.

5. Fournier: Quoted by Futcher.

6. Futcher, T. B.: Diabetes Insipidus, in Osler, W., and McCrae, T.: Modern Medicine, its Theory and Practice, Philadelphia, Lea & Febiger, 1914, ii, 721–728.

7. Gibson, R. B., and Martin, F. T.: The Administration of Pituitary Extract and Histamine in a Case of Diabetes Insipidus, Arch. Int. Med., 1921, xxvii, 351–360.

8. Hoppe-Seyler, G.: Ueber die Beziehung des Diabetes Insipidus zur Hypophyse und seine Behandlung mit Hypophysenextrakt, München. med. Wchnschr., 1915, lxii, 1633–1635.

9. Magnus, R.: Ueber die Enstehung der Hautödeme bei experimenteller hydrämischer Plethora, Arch. f. exper. Path. u. Pharmakol., 1899, xlii, 250–282.

10. Magnus, R., and Schäfer, E. A.: The Action of Pituitary Extracts upon the Kidney, Jour. Physiol., 1901–1902, xxvii, Proc. ix–x.

11. Marshall, E. K., Jr., and Kolls, A. C.: Studies on the Nervous Control of the Kidney in Relation to Diuresis and Urinary Secretion, Am. Jour. Physiol., 1919, xlix, 302–316.

12. Miller, J. L., and Williams, J. L.: The Effect on Blood-pressure and the Non-protein Nitrogen in the Blood of Excessive Fluid Intake, Am. Jour. Med. Sci., 1921, clxi, 327–334.

13. Oppenheim, H.: Die syphilitischen Erkrankungen des Gehirns, specielle Pathologie und Therapie, Vienna, Nothnagel, 1896, ix, 196.

14. Rees, M. H.: The Influence of Pituitary Extracts on the Absorption of Water from the Small Intestine, Am. Jour. Physiol., 1920, liii, 43–48.

15. Rosenbloom, J., and Price, H. T.: A Metabolism Study of a Case of Diabetes Insipidus, Am. Jour. Dis. Child., 1916, xii, 53–60.

16. Rowntree, L. G.: Diabetes Insipidus, in Oxford Medicine, London, Oxford Univ. Press, 1921, iv, 179–193.

17. Schäfer, E. A., and Herring, P. T.: The Action of Pituitary Extracts upon the Kidney, Proc. Roy. Soc. London, 1905–1906, s. B., lxxvii, 571–572.

18. Schäfer, E. A., and Vincent S.: The Physiological Effects of Extracts of the Pituitary Body, Jour. Physiol., 1899 1900, xxv, 87–97.

19. Sollmann, T.: A Manual of Pharmacology and its Applications to Therapeutics and Toxicology, Philadelphia, Saunders, 1917, p. 344.

20. Trousseau, A.: Lectures on Clinical Medicine, London, New Sydenham Soc., 1870, iii, 528–536.

21. Von der Velden, R.: Die Nierenwirkung von Hypophysenextrakten beim Menschen, Berl. klin. Wchnschr., 1913, i, 2083–2086.

DISCUSSION

Dr. D. I. Macht, Baltimore, Md.: In the clinical cases did the doctor note any spasm of the urinary bladder? It is stated that the urinary bladder is more sensitive than the uterus to action of pituitary. It would be interesting to note this.

Dr. Harlow Brooks, New York: Were there any changes in temperature?

Dr. L. G. Rowntree (closing): There was no change in temperature either in the experimental animals or in the clinical cases. We also were interested as to whether contraction of smooth muscle was playing a role. However, we obtain the same effects whether the ureteral catheter was in place, *i. e.*, reaching to the pelvis of the kidney, under which conditions the effect on smooth muscle could not participate in the results. I do not know whether or not there was bladder contraction.

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